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(54) Title: **CELL CYCLE PROGRESSION PROTEINS**

(57) Abstract: Polynucleotides encoding a number of *Drosophila* gene products are provided. Polynucleotide probes derived from these nucleotide sequences, polypeptides encoded by the polynucleotides and antibodies that bind to the polypeptides are also provided.

CELL CYCLE PROGRESSION PROTEINS

The present invention relates to a number of genes implicated in the processes of cell cycle progression, including mitosis and meiosis.

We have now identified a large number of genes in *Drosophila*, mutations in
5 which disrupt cell cycle progression, for example the processes of mitosis and/or meiosis. We have determined the phenotypes of these mutations and recovered nucleotide sequences associated with the corresponding genes. Many of these nucleotide sequences correspond to protein open reading frames (ORFs) present in the *Drosophila* genome.

Accordingly the present invention provides in one aspect a polynucleotide selected
10 from: (a) polynucleotides comprising any one of the nucleotide sequences set out in Examples 1 to 70 or the complement thereof; (b) polynucleotides comprising a nucleotide sequence capable of hybridising to the nucleotide sequences set out in Examples 1 to 70, or a fragment thereof; (c) polynucleotides comprising a nucleotide sequence capable of hybridising to the complement of the nucleotide sequences set out in Examples 1 to 70 or
15 a fragment thereof; (d) polynucleotides comprising a polynucleotide sequence which is degenerate as a result of the genetic code to the polynucleotides defined in (a), (b) or (c).

There is provided, according to another aspect of the present invention, a polynucleotide selected from: (a) polynucleotides comprising any one of the nucleotide sequences set out in Examples 1 to 14 or the complement thereof; (b) polynucleotides
20 comprising a nucleotide sequence capable of hybridising to the nucleotide sequences set out in Examples 1 to 14, or a fragment thereof; (c) polynucleotides comprising a nucleotide sequence capable of hybridising to the complement of the nucleotide sequences set out in Examples 1 to 14 or a fragment thereof; (d) polynucleotides comprising a polynucleotide sequence which is degenerate as a result of the genetic code to the polynucleotides defined
25 in (a), (b) or (c).

We provide, according to yet a further aspect of the present invention, a polynucleotide selected from: (a) polynucleotides comprising any one of the nucleotide

sequences set out in Examples 15 to 19 or the complement thereof; (b) polynucleotides comprising a nucleotide sequence capable of hybridising to the nucleotide sequences set out in Examples 15 to 19, or a fragment thereof; (c) polynucleotides comprising a nucleotide sequence capable of hybridising to the complement of the nucleotide sequences set out in
5 Examples 15 to 19 or a fragment thereof; (d) polynucleotides comprising a polynucleotide sequence which is degenerate as a result of the genetic code to the polynucleotides defined in (a), (b) or (c).

As a further aspect of the present invention, there is provided a polynucleotide selected from: (a) polynucleotides comprising any one of the nucleotide sequences set out
10 in Examples 20 to 30 or the complement thereof; (b) polynucleotides comprising a nucleotide sequence capable of hybridising to the nucleotide sequences set out in Examples 20 to 30, or a fragment thereof; (c) polynucleotides comprising a nucleotide sequence capable of hybridising to the complement of the nucleotide sequences set out in Examples 20 to 30 or a fragment thereof; (d) polynucleotides comprising a polynucleotide sequence
15 which is degenerate as a result of the genetic code to the polynucleotides defined in (a), (b) or (c).

We provide, according to a yet further aspect of the present invention, a polynucleotide selected from: (a) polynucleotides comprising any one of the nucleotide sequences set out in Examples 31 to 53 or the complement thereof; (b) polynucleotides
20 comprising a nucleotide sequence capable of hybridising to the nucleotide sequences set out in Examples 31 to 53, or a fragment thereof; (c) polynucleotides comprising a nucleotide sequence capable of hybridising to the complement of the nucleotide sequences set out in 31 to 53 or a fragment thereof; (d) polynucleotides comprising a polynucleotide sequence which is degenerate as a result of the genetic code to the polynucleotides defined in (a),
25 (b) or (c).

The present invention, in a further aspect, provides a polynucleotide selected from: (a) polynucleotides comprising any one of the nucleotide sequences set out in 54 to 70 or the complement thereof; (b) polynucleotides comprising a nucleotide sequence capable of hybridising to the nucleotide sequences set out in 54 to 70, or a fragment thereof; (c)

polynucleotides comprising a nucleotide sequence capable of hybridising to the complement of the nucleotide sequences set out in 54 to 70 or a fragment thereof; (d) polynucleotides comprising a polynucleotide sequence which is degenerate as a result of the genetic code to the polynucleotides defined in (a), (b) or (c).

- 5 A polynucleotide probe which comprises a fragment of at least 15 nucleotides of a polynucleotide according to any of the above aspects of the invention.

 The present invention also provides a polypeptide which comprises any one of the amino acid sequences set out in Examples 1 to 70 or in any of Examples 1 to 14, Examples 15 to 19, Examples 20 to 30, Examples 31 to 53 and Examples 54 to 70, or a
10 homologue, variant, derivative or fragment thereof.

 Preferably the polypeptide is encoded by a cDNA sequence obtainable from a eukaryotic cDNA library, preferably a metazoan cDNA library (such as insect or mammalian) said DNA sequence comprising a DNA sequence being selectively detectable with a *Drosophila* nucleotide sequence as shown in any one of Examples 1 to 70.

- 15 The term "selectively detectable" means that the cDNA used as a probe is used under conditions where a target cDNA of the invention is found to hybridize to the probe at a level significantly above background. The background hybridization may occur because of other cDNAs present in the cDNA library. In this event background implies a level of signal generated by interaction between the probe and a non-specific cDNA
20 member of the library which is less than 10 fold, preferably less than 100 fold as intense as the specific interaction observed with the target cDNA. The intensity of interaction may be measured, for example, by radiolabelling the probe, e.g. with ³²P. Suitable conditions may be found by reference to the Examples, as well as in the detailed description below.

 A polynucleotide encoding a polypeptide of the invention is also provided.

- 25 The present invention further provides a vector comprising a polynucleotide of the invention, for example an expression vector comprising a polynucleotide of the invention

operably linked to a regulatory sequence capable of directing expression of said polynucleotide in a host cell.

Also provided is an antibody capable of binding a polypeptide of the invention.

5 In a further aspect the present invention provides a method for detecting the presence or absence of a polynucleotide of the invention in a biological sample which method comprises: (a) bringing the biological sample containing DNA or RNA into contact with a probe comprising a nucleotide of the invention under hybridising conditions; and (b) detecting any duplex formed between the probe and nucleic acid in the sample.

10 In another aspect the invention provides a method for detecting a polypeptide of the invention present in a biological sample which comprises: (a) providing an antibody of the invention; (b) incubating a biological sample with said antibody under conditions which allow for the formation of an antibody-antigen complex; and (c) determining whether antibody-antigen complex comprising said antibody is formed.

15 Knowledge of the genes involved in cell cycle progression allows the development of therapeutic agents for the treatment of medical conditions associated with aberrant cell cycle progression. Accordingly, the present invention provides a polynucleotide of the invention for use in therapy. The present invention also provides a polypeptide of the invention for use in therapy. The present invention further provides an antibody of the
20 invention for use in therapy.

In a specific embodiment, the present invention provides a method of treating a tumour or a patient suffering from a proliferative disease, comprising administering to a patient in need of treatment an effective amount of a polynucleotide, polypeptide and/or antibody of the invention.

25 The present invention also provides the use of a polypeptide of the invention in a method of identifying a substance capable of affecting the function of the corresponding

gene. For example, in one embodiment the present invention provides the use of a polypeptide of the invention in an assay for identifying a substance capable of inhibiting cell cycle progression. The substance may inhibit any of the steps or stages in the cell cycle, for example, formation of the nuclear envelope, exit from the quiescent phase of the cell cycle (G0), G1 progression, chromosome decondensation, nuclear envelope breakdown, START, initiation of DNA replication, progression of DNA replication, termination of DNA replication, centrosome duplication, G2 progression, activation of mitotic or meiotic functions, chromosome condensation, centrosome separation, microtubule nucleation, spindle formation and function, interactions with microtubule motor proteins, chromatid separation and segregation, inactivation of mitotic functions, formation of contractile ring, and cytokinesis functions. For example, possible functions of genes of the invention for which it may be desired to identify substances which affect such functions include chromatin binding, formation of replication complexes, replication licensing, phosphorylation or other secondary modification activity, proteolytic degradation, microtubule binding, actin binding, septin binding, microtubule organising centre nucleation activity and binding to components of cell cycle signalling pathways.

In a further aspect the present invention provides a method for identifying a substance capable of binding to a polypeptide of the invention, which method comprises incubating the polypeptide with a candidate substance under suitable conditions and determining whether the substance binds to the polypeptide.

In an additional aspect, the invention provides kits comprising polynucleotides, polypeptides or antibodies of the invention and methods of using such kits in diagnosing the presence of absence of polynucleotides and polypeptides of the invention including deleterious mutant forms.

Also provided is a substance identified by the above methods of the invention. Such substances may be used in a method of therapy, such as in a method of affecting cell cycle progression, for example mitosis and/or meiosis.

The invention also provides a process comprising the steps of: (a) performing one of the above methods; and (b) preparing a quantity of those one or more substances identified as being capable of binding to a polypeptide of the invention.

Also provided is a process comprising the steps of: (a) performing one of the above
5 methods; and (b) preparing a pharmaceutical composition comprising one or more substances identified as being capable of binding to a polypeptide of the invention.

We further provide a method for identifying a substance capable of modulating the function of a polypeptide of the invention or a polypeptide encoded by a polynucleotide of the invention, the method comprising the steps of: incubating the polypeptide with a
10 candidate substance and determining whether activity of the polypeptide is thereby modulated.

A substance identified by a method or assay according to any of the above methods or processes is also provided, as is the use of such a substance in a method of inhibiting the function of a polypeptide. Use of such a substance in a method of regulating a cell
15 division cycle function is also provided.

DETAILED DESCRIPTION OF THE INVENTION

The practice of the present invention will employ, unless otherwise indicated, conventional techniques of chemistry, molecular biology, microbiology, recombinant DNA and immunology, which are within the capabilities of a person of ordinary skill in
20 the art. Such techniques are explained in the literature. See, for example, J. Sambrook, E. F. Fritsch, and T. Maniatis, 1989, *Molecular Cloning: A Laboratory Manual*, Second Edition, Books 1-3, Cold Spring Harbor Laboratory Press; Ausubel, F. M. et al. (1995 and periodic supplements; *Current Protocols in Molecular Biology*, ch. 9, 13, and 16, John Wiley & Sons, New York, N.Y.); B. Roe, J. Crabtree, and A. Kahn, 1996, *DNA Isolation
25 and Sequencing: Essential Techniques*, John Wiley & Sons; J. M. Polak and James O'D. McGee, 1990, *In Situ Hybridization: Principles and Practice*; Oxford University Press; M. J. Gait (Editor), 1984, *Oligonucleotide Synthesis: A Practical Approach*, Irl Press; and, D.

M. J. Lilley and J. E. Dahlberg, 1992, *Methods of Enzymology: DNA Structure Part A: Synthesis and Physical Analysis of DNA* Methods in Enzymology, Academic Press. Each of these general texts is herein incorporated by reference.

Preferably, the polypeptides and polynucleotides of the invention are such that they
 5 give rise to or are associated with defined phenotypes when mutated.

For example, mutations in the polypeptides and polynucleotides of the invention may be associated with a failure to complete cytokinesis; such polypeptides and polynucleotides are conveniently categorised as "Category 1". Phenotypes associated with Category 1 polypeptides and polynucleotides include any one or more of the following,
 10 singly or in combination: Mitotic defects in brain: cytokinesis defect (polyploidy); Male semi-sterile, Meiotic defects in testis: cytokinesis defects, segregation defects.(Seg-01/62); Meiotic defects in testis: cytokinesis defects, abnormal spindles.(Ab-02/12); Mitotic defects in brain: cytokinesis defect (no overcondensation of diploids, high polyploidy); Meiotic defects in testis: cytokinesis defects. Dark bands in eyes, dominant; Meiotic
 15 defects in testis: cytokinesis defects; Meiotic defects in testis:segregation defect, cytokinesis defect(Ck-09/32); Mitotic defects in brain: cytokinesis defect (no overcondensation of diploids, very high polyploidy); Mitotic defects in brain: cytokinesis defect(very high polyploidy); Mitotic defects in brain: cytokinesis defect. Meiotic defects in testis: cytokinesis defects (Mitotic higher level of condensation, polyploidy, Meiotic:
 20 Ck05/07); Mitotic defects in brain, Cytokinesis defect (no overcondensation of diploids, high polyploidy); Mitotic defects in brain: cytokinesis defect (very high polyploidy, chromosomes entangled?); Mitotic defects in brain: cytokinesis defect (very high polyploidy; Meiotic defects in testis: cytokinesis defects (Ck-04/06) `; Female sterile (anaphase bridges, lagging chromosomes); Mitotic defects in brain: cytokinesis defect.
 25 Meiotic defects in testis: cytokinesis defects:(mitotic: high polyploidy, no diploids, higher mitotic index, meiotic: Ck-01/05); Meiotic defects in testis: cytokinesis defects; Meiotic defects in testis: cytokinesis defects(Ck-06/09); Meiotic defects in testis: segregation defects, cytokinesis defect(Ck-07/35); Meiotic defects in testis: cytokinesis defects.

Alternatively, mutations in the polypeptides and polynucleotides of the invention may be associated with a failure to enter M-phase; such polypeptides and polynucleotides are conveniently categorised as "Category 2". Phenotypes associated with Category 2 polypeptides and polynucleotides include any one or more of the following, singly or in combination: Meiotic defects in testis: no division(no meiosis); Mitotic defects in brain: no mitosis; Meiotic defects in testis: segregation defects, meiotic failure(Mf-07/75); Meiotic defects in testis: segregation defects, meiotic failure(Mf-05/31); Meiotic defects in testis: cytokinesis defects, meiotic failure(Mf-02/15).

Mutations in the polypeptides and polynucleotides of the invention may be associated with a metaphase arrest phenotype ("Category 3"). Phenotypes associated with Category 3 polypeptides and polynucleotides include any one or more of the following, singly or in combination: Mitotic defects in brain: prometaphase arrest (overcondensation, polyploidy, scattered chromosomes with bipolar spindle); Male sterile, Female sterile, Mitotic defects in brain: prometaphase arrest (Overcondensation, polyploidy, fewer anaphases, high mitotic index, scattered chromosomes with bipolar spindle); Mitotic defects in brain: (weak overcondensation, metaphase with bipolar spindle); Mitotic defects in brain: prometaphase arrest; Mitotic defects in brain: metaphase arrest; Mitotic defects in brain: metaphase arrest. (overcondensation, polyploidy, aneuploidy, few anaphases, high mitotic index, metaphase with bent bipolar spindle); Mitotic defects in brain: metaphase arrest.(overcondensation, polyploidy, few anaphases, high mitotic index, metaphase with bent bipolar spindle); Mitotic defects in brain: Metaphase arrest (overcondensation, polyploidy, aneuploidy, no anaphases, high mitotic index, metaphase with bipolar spindle); Mitotic defects in brain: metaphase arrest (overcondensation, metaphase with bipolar spindle; Meiotic defects in testis: segregation defects, multipolar spindles (Mul-02/29); Meiotic defects in testis: cytokinesis defects, abnormal spindles.(Ab-01/03); Mitotic defects in brain: metaphase arrest; Mitotic defects in brain: metaphase arrest (overcondensation, polyploidy, metaphase with bipolar spindle); Mitotic defects in brain: metaphase arrest. Meiotic defects in testis: segregation defects. Abnormal spindles (mitotic: High mitotic index, meiotic: Ab-08/24); Mitotic defects in brain: metaphase arrest(overcondensation, few anaphases, some polyploids); Mitotic defects in brain: prometaphase arrest (overcondensation, fewer anaphases, metaphase with bipolar spindle);

Mitotic defects in brain: metaphase arrest(condensation, no polyploidy, no anaphases, metaphase with bipolar spindle).

Mutations in Category 4 polypeptides and polynucleotides of the invention may be associated with an anaphase defect phenotype; phenotypes associated with Category 4

5 polypeptides and polynucleotides include any one or more of the following, singly or in combination: Mitotic defects in brain: anaphase defects (overcondensation, high polyploidy, some lagging chromosomes); Meiotic defects in testis: segregation defects; Male and female sterile, small wings, meiotic defects in testis: segregation defects, elongation defect; Mitotic defects in brain: anaphase defects(overcondensation, anaphase

10 bridge, metaphase with swollen chromosomes and bipolar spindle); Mitotic defects in brain: Anaphase defects. (overcondensation, aneuploidy, some lagging chromosomes and breaks); Meiotic defects in testis: segregation defects; Meiotic defects in testis: segregation defects, multi-stage defects (Pl-02/17); Meiotic defects in testis: segregation defects, multi-stage defects (Pl-02/18); Meiotic defects in testis: cytokinesis defects,

15 segregation defects (seg-01/01); Mitotic defects in brain: cytokinesis defect. Meiotic defects in testis: cytokinesis defect. Multi-stage defects Polyploidy, no overcondensation Pl-01/10; Meiotic defects in testis: segregation defects, abnormal spindles. (Ab-03/30); Mitotic defects in brain: anaphase defects (weak, higher condensation, some polyploidy, fewer anaphases, polyploids with monopolar spindles); Mitotic defects in brain: anaphase

20 defects (overcondensation, polyploidy (with overcondensation), few anaphases, metaphase with bipolar spindle); Meiotic defects in testis: cytokinesis defects; Meiotic defects in testis: segregation defects,multipolar spindles(Mul-02/22); Meiotic defects in testis: segregation defects, abnormal spindles (Ab-04/26); Meiotic defects in testis: cytokinesis defects,abnormal spindles (Ab-16/13); Mitotic defects in brain: anaphase defects. Meiotic

25 defects in testis: segregation defects, abnormal spindles (mitotic : Overcondensation, lagging chromosomes/less aligned metaphase with bipolar spindles, Meiotic: Ab-06/20); Meiotic defects in testis: segregation defects; Meiotic defects in testis: no division (no meiosis); Meiotic defects in testis: segregation defects, abnormal spindles (Ab-12/48); Meiotic defects in testis: segregation defects, multipolar spindles(mitotic: High polyploids,

30 no diploids, higher mitotic index Meiotic: Mul-02/59); Meiotic defects in testis: segregation defect; Meiotic defects in testis: segregation defects,abnormal spindles

- (meiotic: Ab-08/42); Female sterile. Meiotic defects in testis: cytokinesis defects, segregation defects (Mitotic: Less condensed chromosomes, nuclear bridges, Meiotic: Seg-01/02; Mitotic defects in brain: anaphase defects; Meiotic defects in testis: cytokinesis defects, abnormal spindles (Ab-01/04); Meiotic defects in testis: segregation
- 5 defects (overcondensation, fewer anaphases); Mitotic defects in brain: (some overcondensation, anaphase bridge, metaphase with swollen chromosome and bipolar spindle).

- A fifth category ("Category 5") of polypeptides and polynucleotides of the invention are associated with the presence of small imaginal discs (block to proliferation).
- 10 Phenotypes associated with Category 5 polypeptides and polynucleotides include any one or more of the following, singly or in combination: 2nd chromosome, small imaginal discs.

The polypeptides and polynucleotides of the invention may also be categorised according to their function, or their putative function.

- 15 For example, the polypeptides described here preferably comprise, and the polynucleotides described here are ones which preferably encode polypeptides comprising, any one or more of the following: a CBP activator protein; a CCR4-associated regulator of polymerase II transcription; a CTP synthase (CTPS); a Cyclin specific ubiquitin conjugating enzyme; a DNA packaging protein; a DNA repair protein; a DNA-
- 20 binding protein involved in chromosomal organisation; a DNase IV; a EIF4G2 translation initiation factor; a eukaryotic translation initiation factor 6; a Ecdysone-induced protein 78C; a Egf2 translation factor; a G protein-coupled receptor kinase 7; a GTPase exchange factor; a phosphatidylinositol transfer protein beta isoform; a His-rich protein; a Lk6 kinase; a MAP kinase; a MAP kinase interacting kinase 1; a N-arginine dibasic
- 25 convertase; a Phosphatidylinositol transfer protein; a RIP protein kinase; a RNA binding motif, single stranded interacting protein; a RNA binding protein; a RYK receptor tyrosine kinase; a Ribosomal protein L1; a selenide, water dikinase 1; a selenium donor protein 1; a selenophosphate synthetase 1; a Sqv-7-like protein; a sugar modification protein; a protein involved in cytokinesis and signalling; a TEK tyrosine kinase; a Translation elongation

factor; a UDP-galactose transporter; a v-erba related protein; a WD40 protein; a brahma protein; a calcium binding protein; a cell adhesion protein; a chaperone; a chromodomain helicase DNA binding protein; a chromodomain-helicase-DNA-binding protein; a coiled coil protein with ubiquitin like domain; a component of the 19S proteasome regulatory particle; a couch potato RNA binding protein; a cytidine 5-prime triphosphate synthetase; a cytoskeletal structural protein; a death domain containing protein; a developmentally expressed in axons of the CNS; a diacylglycerol-activated/phospholipid dependent protein kinase C inhibitor; a diazepam binding inhibitor; a diphosphate kinase; a dodecasatellite DNA binding protein; a doughnut protein tyrosine kinase; an elongation factor 2; a
10 endoplasmic reticulum ATPase; a eukaryotic translation initiation factor 4E binding protein 2; a factor involved in axon guidance; a fatty-acid-Coenzyme A ligase; a flap structure-specific endonuclease 1; a protein involved in the formation of the contractile ring and the initiation of cytokinesis; a glucose-6-phosphate transporter; a glycoprotein glucosyltransferase; a growth factor; a transmembrane receptor protein tyrosine kinase
15 involved in cell growth and maintenance; a guanyl-nucleotide exchange factor involved in signal transduction; a heat shock protein; a helicase; a high density lipoprotein binding protein; a histone acetyl transferase transcriptional activator; a histone acetyltransferase; a histone acetyltransferase GCN5; a protein involved in development of the abdomen (embryos); a protein involved in the development of the imaginal discs (larvae or pupae);
20 a kinesin like protein 67a; a ligand-dependent nuclear receptor; a ligand-dependent nuclear receptor; a lola-like specific RNA polymerase II transcription factor; a matrix associated protein; a membrane glycoprotein; a mitotic heterochromatin fragment clone CH(2)6; a motor protein; a motor protein involved in cytoskeleton organization; a mushroom body RNA binding protein; a myosin like proteins; a nemo-like kinase; a non-ATPase protein; a
25 nuclear receptor NR1E1; a nucleic acid binding protein; a nucleoside diphosphate kinase (NBR-A); a oly(rC)-binding protein 2 (hnRNP-E1); a peroxisome biogenesis factor 1; a phospholipid transporter involved in lipid metabolism; a phosphatase or enhancer of Pi uptake protein; a protease; a proteasome regulatory particle; a protein involved in cytoskeleton organization and/or biogenesis; a protein kinase associated with
30 microtubules; a protein kinase mitogen-activated 7; a protein serine/threonine kinase involved in cell cycle, possibly targeted to cytoskeleton; a protein serine/threonine kinase involved in eye morphogenesis; a protein which associates with cdc25 phosphatase; a

protein which induces apoptosis; a ribonuclease P; a ribonuclease P protein subunit p29; a ser/thr phosphatase; a signal transduction protein; a signal transport protein; a sin3-associated polypeptide; a single stranded DNA/RNA binding protein; a sodium-dependent dicarboxylate transporters; a ssDNA/RNA binding proteins; a striatin, calmodulin-binding protein (STRN); a structural protein of ribosome involved in protein biosynthesis; a
 5 subtelomeric heterochromatin repeats; a sugar acetylase; a sugar modification protein; a suppressor of ras; a tRNA processing enzyme Ribonuclease P protein subunit; a thyroid hormone responsive gene; a tie receptor protein tyrosine kinase; a transacylase; a transcription factor; a transcription factor involved in chromatin remodelling; a
 10 transcriptional regulation of c-myc expression; a transcriptional regulator; a transcriptional regulators/telomeric silencing; a translation initiation factor; a tumor metastasis inhibitor; a tyrosine 3-monooxygenase/tryptophan 5-monooxygenase activation protein; a ubiquitin carrier protein; a ubiquitin-conjugating enzyme; a ugtUDP-glucose-glycoprotein glucosyltransferase; a zinc finger protein; an RNA polymerase II transcription factor; an
 15 acetylcholinesterase (YT blood group) precursor; an actin binding protein; an actin dependent regulator of chromatin; an acyl-CoA-binding protein; an alanine:glyoxylate aminotransferase; an alpha esterase; an ankyrin protein; an imitation-SWI protein; and an integrin beta 4 binding protein.

POLYPEPTIDES

20 It will be understood that polypeptides of the invention are not limited to polypeptides having the amino acid sequence set out in Examples 1 to 70 or fragments thereof but also include homologous sequences obtained from any source, for example related viral/bacterial proteins, cellular homologues and synthetic peptides, as well as variants or derivatives thereof.

25 Thus polypeptides of the invention also include those encoding homologues from other species including animals such as mammals (e.g. mice, rats or rabbits), especially primates, more especially humans. More specifically, homologues included within the scope of the invention include human homologues.

Thus, the present invention covers variants, homologues or derivatives of the amino acid sequence set out in Examples 1 to 70, as well as variants, homologues or derivatives of the nucleotide sequence coding for the amino acid sequences of the present invention.

5 In the context of the present invention, a homologous sequence is taken to include an amino acid sequence which is at least 15, 20, 25, 30, 40, 50, 60, 70, 80 or 90% identical, preferably at least 95 or 98% identical at the amino acid level over at least 50 or 100, preferably 200, 300, 400 or 500 amino acids with any one of the polypeptide sequences shown in the Examples. In particular, homology should typically be considered
10 with respect to those regions of the sequence known to be essential for protein function rather than non-essential neighbouring sequences. This is especially important when considering homologous sequences from distantly related organisms.

Although homology can also be considered in terms of similarity (i.e. amino acid residues having similar chemical properties/functions), in the context of the present
15 invention it is preferred to express homology in terms of sequence identity.

Homology comparisons can be conducted by eye, or more usually, with the aid of readily available sequence comparison programs. These publicly and commercially available computer programs can calculate % homology between two or more sequences.

% homology may be calculated over contiguous sequences, i.e. one sequence is
20 aligned with the other sequence and each amino acid in one sequence directly compared with the corresponding amino acid in the other sequence, one residue at a time. This is called an "ungapped" alignment. Typically, such ungapped alignments are performed only over a relatively short number of residues (for example less than 50 contiguous amino acids).

25 Although this is a very simple and consistent method, it fails to take into consideration that, for example, in an otherwise identical pair of sequences, one insertion or deletion will cause the following amino acid residues to be put out of alignment, thus

potentially resulting in a large reduction in % homology when a global alignment is performed. Consequently, most sequence comparison methods are designed to produce optimal alignments that take into consideration possible insertions and deletions without penalising unduly the overall homology score. This is achieved by inserting "gaps" in the sequence alignment to try to maximise local homology.

However, these more complex methods assign "gap penalties" to each gap that occurs in the alignment so that, for the same number of identical amino acids, a sequence alignment with as few gaps as possible - reflecting higher relatedness between the two compared sequences - will achieve a higher score than one with many gaps. "Affine gap costs" are typically used that charge a relatively high cost for the existence of a gap and a smaller penalty for each subsequent residue in the gap. This is the most commonly used gap scoring system. High gap penalties will of course produce optimised alignments with fewer gaps. Most alignment programs allow the gap penalties to be modified. However, it is preferred to use the default values when using such software for sequence comparisons. For example when using the GCG Wisconsin Bestfit package (see below) the default gap penalty for amino acid sequences is -12 for a gap and -4 for each extension.

Calculation of maximum % homology therefore firstly requires the production of an optimal alignment, taking into consideration gap penalties. A suitable computer program for carrying out such an alignment is the GCG Wisconsin Bestfit package (University of Wisconsin, U.S.A; Devereux *et al.*, 1984, Nucleic Acids Research 12:387). Examples of other software than can perform sequence comparisons include, but are not limited to, the BLAST package (see Ausubel *et al.*, 1999 *ibid* - Chapter 18), FASTA (Atschul *et al.*, 1990, J. Mol. Biol., 403-410) and the GENWORKS suite of comparison tools. Both BLAST and FASTA are available for offline and online searching (see Ausubel *et al.*, 1999 *ibid*, pages 7-58 to 7-60). However it is preferred to use the GCG Bestfit program.

Although the final % homology can be measured in terms of identity, the alignment process itself is typically not based on an all-or-nothing pair comparison. Instead, a scaled similarity score matrix is generally used that assigns scores to each

pairwise comparison based on chemical similarity or evolutionary distance. An example of such a matrix commonly used is the BLOSUM62 matrix - the default matrix for the BLAST suite of programs. GCG Wisconsin programs generally use either the public default values or a custom symbol comparison table if supplied (see user manual for
5 further details). It is preferred to use the public default values for the GCG package, or in the case of other software, the default matrix, such as BLOSUM62.

Once the software has produced an optimal alignment, it is possible to calculate % homology, preferably % sequence identity. The software typically does this as part of the sequence comparison and generates a numerical result.

10 The terms "variant" or "derivative" in relation to the amino acid sequences of the present invention includes any substitution of, variation of, modification of, replacement of, deletion of or addition of one (or more) amino acids from or to the sequence providing the resultant amino acid sequence retains substantially the same activity as the unmodified sequence, preferably having at least the same activity as the polypeptides presented in the
15 sequence listings in the Examples.

Polypeptides having the amino acid sequence shown in the Examples, or fragments or homologues thereof may be modified for use in the present invention. Typically, modifications are made that maintain the biological activity of the sequence. Amino acid substitutions may be made, for example from 1, 2 or 3 to 10, 20 or 30 substitutions
20 provided that the modified sequence retains the biological activity of the unmodified sequence. Alternatively, modifications may be made to deliberately inactivate one or more functional domains of the polypeptides of the invention. Amino acid substitutions may include the use of non-naturally occurring analogues, for example to increase blood plasma half-life of a therapeutically administered polypeptide.

25 Conservative substitutions may be made, for example according to the Table below. Amino acids in the same block in the second column and preferably in the same line in the third column may be substituted for each other:

ALIPHATIC	Non-polar	G A P
		I L V
	Polar - uncharged	C S T M
		N Q
	Polar - charged	D E
		K R
AROMATIC		H F W Y

Polypeptides of the invention also include fragments of the full length sequences mentioned above. Preferably said fragments comprise at least one epitope. Methods of identifying epitopes are well known in the art. Fragments will typically comprise at least 6 amino acids, more preferably at least 10, 20, 30, 50 or 100 amino acids.

- 5 Proteins of the invention are typically made by recombinant means, for example as described below. However they may also be made by synthetic means using techniques well known to skilled persons such as solid phase synthesis. Proteins of the invention may also be produced as fusion proteins, for example to aid in extraction and purification. Examples of fusion protein partners include glutathione-S-transferase (GST), 6xHis, GAL4 (DNA binding and/or transcriptional activation domains) and β -galactosidase. It may also be convenient to include a proteolytic cleavage site between the fusion protein partner and the protein sequence of interest to allow removal of fusion protein sequences. Preferably the fusion protein will not hinder the function of the protein of interest sequence. Proteins of the invention may also be obtained by purification of cell extracts from animal cells.
- 10
- 15

- Proteins of the invention may be in a substantially isolated form. It will be understood that the protein may be mixed with carriers or diluents which will not interfere with the intended purpose of the protein and still be regarded as substantially isolated. A protein of the invention may also be in a substantially purified form, in which case it will generally comprise the protein in a preparation in which more than 90%, e.g. 95%, 98% or 99% of the protein in the preparation is a protein of the invention.
- 20

A polypeptide of the invention may be labeled with a revealing label. The revealing label may be any suitable label which allows the polypeptide to be detected. Suitable labels include radioisotopes, e.g. ^{125}I , enzymes, antibodies, polynucleotides and linkers such as biotin. Labeled polypeptides of the invention may be used in diagnostic procedures such as
5 immunoassays to determine the amount of a polypeptide of the invention in a sample. Polypeptides or labeled polypeptides of the invention may also be used in serological or cell-mediated immune assays for the detection of immune reactivity to said polypeptides in animals and humans using standard protocols.

A polypeptide or labeled polypeptide of the invention or fragment thereof may also
10 be fixed to a solid phase, for example the surface of an immunoassay well or dipstick. Such labeled and/or immobilised polypeptides may be packaged into kits in a suitable container along with suitable reagents, controls, instructions and the like. Such polypeptides and kits may be used in methods of detection of antibodies to the polypeptides or their allelic or species variants by immunoassay.

15 Immunoassay methods are well known in the art and will generally comprise: (a) providing a polypeptide comprising an epitope bindable by an antibody against said protein; (b) incubating a biological sample with said polypeptide under conditions which allow for the formation of an antibody-antigen complex; and (c) determining whether antibody-antigen complex comprising said polypeptide is formed.

20 Polypeptides of the invention may be used in *in vitro* or *in vivo* cell culture systems to study the role of their corresponding genes and homologues thereof in cell function, including their function in disease. For example, truncated or modified polypeptides may be introduced into a cell to disrupt the normal functions which occur in the cell. The polypeptides of the invention may be introduced into the cell by *in situ* expression of the
25 polypeptide from a recombinant expression vector (see below). The expression vector optionally carries an inducible promoter to control the expression of the polypeptide.

The use of appropriate host cells, such as insect cells or mammalian cells, is expected to provide for such post-translational modifications (e.g. myristolation,

glycosylation, truncation, lapidation and tyrosine, serine or threonine phosphorylation) as may be needed to confer optimal biological activity on recombinant expression products of the invention. Such cell culture systems in which polypeptides of the invention are expressed may be used in assay systems to identify candidate substances which interfere
5 with or enhance the functions of the polypeptides of the invention in the cell.

POLYNUCLEOTIDES

Polynucleotides of the invention include polynucleotides that comprise any one or more of the nucleic acid sequences set out in Examples 1 to 70 and fragments thereof. Polynucleotides of the invention also include polynucleotides encoding the polypeptides
10 of the invention. It will be understood by a skilled person that numerous different polynucleotides can encode the same polypeptide as a result of the degeneracy of the genetic code. In addition, it is to be understood that skilled persons may, using routine techniques, make nucleotide substitutions that do not affect the polypeptide sequence encoded by the polynucleotides of the invention to reflect the codon usage of any
15 particular host organism in which the polypeptides of the invention are to be expressed.

Polynucleotides of the invention may comprise DNA or RNA. They may be single-stranded or double-stranded. They may also be polynucleotides which include within them synthetic or modified nucleotides. A number of different types of modification to oligonucleotides are known in the art. These include methylphosphonate
20 and phosphorothioate backbones, addition of acridine or polylysine chains at the 3' and/or 5' ends of the molecule. For the purposes of the present invention, it is to be understood that the polynucleotides described herein may be modified by any method available in the art. Such modifications may be carried out in order to enhance the *in vivo* activity or life span of polynucleotides of the invention.

25 The terms "variant", "homologue" or "derivative" in relation to the nucleotide sequence of the present invention include any substitution of, variation of, modification of, replacement of, deletion of or addition of one (or more) nucleic acid from or to the

sequence. Preferably said variant, homologues or derivatives code for a polypeptide having biological activity.

As indicated above, with respect to sequence homology, preferably there is at least 50 or 75%, more preferably at least 85%, more preferably at least 90% homology to the sequences shown in the sequence listing herein. More preferably there is at least 95%,
5 more preferably at least 98%, homology. Nucleotide homology comparisons may be conducted as described above. A preferred sequence comparison program is the GCG Wisconsin Bestfit program described above. The default scoring matrix has a match value of 10 for each identical nucleotide and -9 for each mismatch. The default gap creation
10 penalty is -50 and the default gap extension penalty is -3 for each nucleotide.

The present invention also encompasses nucleotide sequences that are capable of hybridising selectively to the sequences presented herein, or any variant, fragment or derivative thereof, or to the complement of any of the above. Nucleotide sequences are preferably at least 15 nucleotides in length, more preferably at least 20, 30, 40 or 50
15 nucleotides in length.

The term "hybridization" as used herein shall include "the process by which a strand of nucleic acid joins with a complementary strand through base pairing" as well as the process of amplification as carried out in polymerase chain reaction technologies.

Polynucleotides of the invention capable of selectively hybridising to the nucleotide sequences presented herein, or to their complement, will be generally at least
20 70%, preferably at least 80 or 90% and more preferably at least 95% or 98% homologous to the corresponding nucleotide sequences presented herein over a region of at least 20, preferably at least 25 or 30, for instance at least 40, 60 or 100 or more contiguous nucleotides.

25 The term "selectively hybridizable" means that the polynucleotide used as a probe is used under conditions where a target polynucleotide of the invention is found to hybridize to the probe at a level significantly above background. The background

hybridization may occur because of other polynucleotides present, for example, in the cDNA or genomic DNA library being screening. In this event, background implies a level of signal generated by interaction between the probe and a non-specific DNA member of the library which is less than 10 fold, preferably less than 100 fold as intense as the
5 specific interaction observed with the target DNA. The intensity of interaction may be measured, for example, by radiolabelling the probe, e.g. with ^{32}P .

Hybridization conditions are based on the melting temperature (T_m) of the nucleic acid binding complex, as taught in Berger and Kimmel (1987, Guide to Molecular Cloning Techniques, Methods in Enzymology, Vol 152, Academic Press, San Diego CA), and
10 confer a defined "stringency" as explained below.

Maximum stringency typically occurs at about $T_m - 5^\circ\text{C}$ (5°C below the T_m of the probe); high stringency at about 5°C to 10°C below T_m ; intermediate stringency at about 10°C to 20°C below T_m ; and low stringency at about 20°C to 25°C below T_m . As will be understood by those of skill in the art, a maximum stringency hybridization can be used to
15 identify or detect identical polynucleotide sequences while an intermediate (or low) stringency hybridization can be used to identify or detect similar or related polynucleotide sequences.

In a preferred aspect, the present invention covers nucleotide sequences that can hybridise to the nucleotide sequence of the present invention under stringent conditions
20 (e.g. 65°C and $0.1\times\text{SSC}$ ($1\times\text{SSC} = 0.15\text{ M NaCl}$, $0.015\text{ M Na}_3\text{ Citrate pH } 7.0$)).

Where the polynucleotide of the invention is double-stranded, both strands of the duplex, either individually or in combination, are encompassed by the present invention. Where the polynucleotide is single-stranded, it is to be understood that the complementary sequence of that polynucleotide is also included within the scope of the present invention.

25 Polynucleotides which are not 100% homologous to the sequences of the present invention but fall within the scope of the invention can be obtained in a number of ways. Other variants of the sequences described herein may be obtained for example by probing

DNA libraries made from a range of individuals, for example individuals from different populations. In addition, other viral/bacterial, or cellular homologues particularly cellular homologues found in mammalian cells (e.g. rat, mouse, bovine and primate cells), may be obtained and such homologues and fragments thereof in general will be capable of

5 selectively hybridising to the sequences shown in the Examples. Such sequences may be obtained by probing cDNA libraries made from or genomic DNA libraries from other animal species, and probing such libraries with probes comprising all or part of any one of the sequences shown in the Examples under conditions of medium to high stringency. The nucleotide sequences of the human homologues described in the Examples, may

10 preferably be used to identify other primate/mammalian homologues since nucleotide homology between human sequences and mammalian sequences is likely to be higher than is the case for the *Drosophila* sequences identified herein.

Similar considerations apply to obtaining species homologues and allelic variants of the polypeptide or nucleotide sequences of the invention.

15 Variants and strain/species homologues may also be obtained using degenerate PCR which will use primers designed to target sequences within the variants and homologues encoding conserved amino acid sequences within the sequences of the present invention. Conserved sequences can be predicted, for example, by aligning the amino acid sequences from several variants/homologues. Sequence alignments can be performed

20 using computer software known in the art. For example the GCG Wisconsin PileUp program is widely used.

The primers used in degenerate PCR will contain one or more degenerate positions and will be used at stringency conditions lower than those used for cloning sequences with single sequence primers against known sequences. It will be appreciated by the skilled

25 person that overall nucleotide homology between sequences from distantly related organisms is likely to be very low and thus in these situations degenerate PCR may be the method of choice rather than screening libraries with labeled fragments the sequences disclosed in the Examples.

In addition, homologous sequences may be identified by searching nucleotide and/or protein databases using search algorithms such as the BLAST suite of programs. This approach is described in the Examples.

Alternatively, such polynucleotides may be obtained by site directed mutagenesis of characterised sequences, such as the sequences disclosed in the Examples. This may be useful where for example silent codon changes are required to sequences to optimise codon preferences for a particular host cell in which the polynucleotide sequences are being expressed. Other sequence changes may be desired in order to introduce restriction enzyme recognition sites, or to alter the property or function of the polypeptides encoded by the polynucleotides. For example, further changes may be desirable to represent particular coding changes found in the sequences disclosed in the Examples which give rise to mutant genes which have lost their regulatory function. Probes based on such changes can be used as diagnostic probes to detect such mutants.

Polynucleotides of the invention may be used to produce a primer, e.g. a PCR primer, a primer for an alternative amplification reaction, a probe e.g. labeled with a revealing label by conventional means using radioactive or non-radioactive labels, or the polynucleotides may be cloned into vectors. Such primers, probes and other fragments will be at least 8, 9, 10, or 15, preferably at least 20, for example at least 25, 30 or 40 nucleotides in length, and are also encompassed by the term polynucleotides of the invention as used herein.

Polynucleotides such as a DNA polynucleotides and probes according to the invention may be produced recombinantly, synthetically, or by any means available to those of skill in the art. They may also be cloned by standard techniques.

In general, primers will be produced by synthetic means, involving a step wise manufacture of the desired nucleic acid sequence one nucleotide at a time. Techniques for accomplishing this using automated techniques are readily available in the art.

Longer polynucleotides will generally be produced using recombinant means, for example using a PCR (polymerase chain reaction) cloning techniques. This will involve making a pair of primers (e.g. of about 15 to 30 nucleotides) flanking a region of the lipid targeting sequence which it is desired to clone, bringing the primers into contact with
5 mRNA or cDNA obtained from an animal or human cell, performing a polymerase chain reaction under conditions which bring about amplification of the desired region, isolating the amplified fragment (e.g. by purifying the reaction mixture on an agarose gel) and recovering the amplified DNA. The primers may be designed to contain suitable restriction enzyme recognition sites so that the amplified DNA can be cloned into a
10 suitable cloning vector

Polynucleotides or primers of the invention may carry a revealing label. Suitable labels include radioisotopes such as ^{32}P or ^{35}S , enzyme labels, or other protein labels such as biotin. Such labels may be added to polynucleotides or primers of the invention and may be detected using by techniques known *per se*.

15 Polynucleotides or primers of the invention or fragments thereof labeled or unlabeled may be used by a person skilled in the art in nucleic acid-based tests for detecting or sequencing polynucleotides of the invention in the human or animal body.

Such tests for detecting generally comprise bringing a biological sample containing DNA or RNA into contact with a probe comprising a polynucleotide or primer of the
20 invention under hybridising conditions and detecting any duplex formed between the probe and nucleic acid in the sample. Such detection may be achieved using techniques such as PCR or by immobilising the probe on a solid support, removing nucleic acid in the sample which is not hybridised to the probe, and then detecting nucleic acid which has hybridised to the probe. Alternatively, the sample nucleic acid may be immobilised on a
25 solid support, and the amount of probe bound to such a support can be detected. Suitable assay methods of this and other formats can be found in for example WO89/03891 and WO90/13667.

Tests for sequencing nucleotides of the invention include bringing a biological sample containing target DNA or RNA into contact with a probe comprising a polynucleotide or primer of the invention under hybridising conditions and determining the sequence by, for example the Sanger dideoxy chain termination method (see
5 Sambrook *et al.*).

Such a method generally comprises elongating, in the presence of suitable reagents, the primer by synthesis of a strand complementary to the target DNA or RNA and selectively terminating the elongation reaction at one or more of an A, C, G or T/U residue; allowing strand elongation and termination reaction to occur; separating out
10 according to size the elongated products to determine the sequence of the nucleotides at which selective termination has occurred. Suitable reagents include a DNA polymerase enzyme, the deoxynucleotides dATP, dCTP, dGTP and dTTP, a buffer and ATP. Dideoxynucleotides are used for selective termination.

Tests for detecting or sequencing nucleotides of the invention in a biological
15 sample may be used to determine particular sequences within cells in individuals who have, or are suspected to have, an altered gene sequence, for example within cancer cells including leukaemia cells and solid tumours such as breast, ovary, lung, colon, pancreas, testes, liver, brain, muscle and bone tumours. Cells from patients suffering from a proliferative disease may also be tested in the same way.

20 In addition, the identification of the genes described in the Examples will allow the role of these genes in hereditary diseases to be investigated. In general, this will involve establishing the status of the gene (e.g. using PCR sequence analysis), in cells derived from animals or humans with, for example, neurological disorders or neoplasms.

The probes of the invention may conveniently be packaged in the form of a test kit
25 in a suitable container. In such kits the probe may be bound to a solid support where the assay format for which the kit is designed requires such binding. The kit may also contain suitable reagents for treating the sample to be probed, hybridising the probe to nucleic acid in the sample, control reagents, instructions, and the like.

NUCLEIC ACID VECTORS

Polynucleotides of the invention can be incorporated into a recombinant replicable vector. The vector may be used to replicate the nucleic acid in a compatible host cell. Thus in a further embodiment, the invention provides a method of making polynucleotides of the invention by introducing a polynucleotide of the invention into a replicable vector,
5 introducing the vector into a compatible host cell, and growing the host cell under conditions which bring about replication of the vector. The vector may be recovered from the host cell. Suitable host cells include bacteria such as *E. coli*, yeast, mammalian cell lines and other eukaryotic cell lines, for example insect Sf9 cells.

10 Preferably, a polynucleotide of the invention in a vector is operably linked to a control sequence that is capable of providing for the expression of the coding sequence by the host cell, i.e. the vector is an expression vector. The term "operably linked" means that the components described are in a relationship permitting them to function in their intended manner. A regulatory sequence "operably linked" to a coding sequence is ligated
15 in such a way that expression of the coding sequence is achieved under condition compatible with the control sequences.

The control sequences may be modified, for example by the addition of further transcriptional regulatory elements to make the level of transcription directed by the control sequences more responsive to transcriptional modulators.

20 Vectors of the invention may be transformed or transfected into a suitable host cell as described below to provide for expression of a protein of the invention. This process may comprise culturing a host cell transformed with an expression vector as described above under conditions to provide for expression by the vector of a coding sequence encoding the protein, and optionally recovering the expressed protein. Vectors will be
25 chosen that are compatible with the host cell used.

The vectors may be for example, plasmid or virus vectors provided with an origin of replication, optionally a promoter for the expression of the said polynucleotide and

optionally a regulator of the promoter. The vectors may contain one or more selectable marker genes, for example an ampicillin resistance gene in the case of a bacterial plasmid or a neomycin resistance gene for a mammalian vector. Vectors may be used, for example, to transfect or transform a host cell.

- 5 Control sequences operably linked to sequences encoding the polypeptide of the invention include promoters/enhancers and other expression regulation signals. These control sequences may be selected to be compatible with the host cell for which the expression vector is designed to be used in. The term promoter is well-known in the art and encompasses nucleic acid regions ranging in size and complexity from minimal
10 promoters to promoters including upstream elements and enhancers.

- The promoter is typically selected from promoters which are functional in mammalian cells, although prokaryotic promoters and promoters functional in other eukaryotic cells, such as insect cells, may be used. The promoter is typically derived from promoter sequences of viral or eukaryotic genes. For example, it may be a promoter
15 derived from the genome of a cell in which expression is to occur. With respect to eukaryotic promoters, they may be promoters that function in a ubiquitous manner (such as promoters of α -actin, β -actin, tubulin) or, alternatively, a tissue-specific manner (such as promoters of the genes for pyruvate kinase). They may also be promoters that respond to specific stimuli, for example promoters that bind steroid hormone receptors. Viral
20 promoters may also be used, for example the Moloney murine leukaemia virus long terminal repeat (MMLV LTR) promoter, the rous sarcoma virus (RSV) LTR promoter or the human cytomegalovirus (CMV) IE promoter.

- It may also be advantageous for the promoters to be inducible so that the levels of expression of the heterologous gene can be regulated during the life-time of the cell.
25 Inducible means that the levels of expression obtained using the promoter can be regulated.

In addition, any of these promoters may be modified by the addition of further regulatory sequences, for example enhancer sequences. Chimeric promoters may also be

used comprising sequence elements from two or more different promoters described above.

Polynucleotides according to the invention may also be inserted into the vectors described above in an antisense orientation to provide for the production of antisense
5 RNA. Antisense RNA or other antisense polynucleotides may also be produced by synthetic means. Such antisense polynucleotides may be used in a method of controlling the levels of RNAs transcribed from genes comprising any one of the polynucleotides of the invention.

HOST CELLS

10 Vectors and polynucleotides of the invention may be introduced into host cells for the purpose of replicating the vectors/polynucleotides and/or expressing the polypeptides of the invention encoded by the polynucleotides of the invention. Although the polypeptides of the invention may be produced using prokaryotic cells as host cells, it is preferred to use eukaryotic cells, for example yeast, insect or mammalian cells, in
15 particular mammalian cells.

Vectors/polynucleotides of the invention may be introduced into suitable host cells using a variety of techniques known in the art, such as transfection, transformation and electroporation. Where vectors/polynucleotides of the invention are to be administered to animals, several techniques are known in the art, for example infection with recombinant
20 viral vectors such as retroviruses, herpes simplex viruses and adenoviruses, direct injection of nucleic acids and biolistic transformation.

PROTEIN EXPRESSION AND PURIFICATION

Host cells comprising polynucleotides of the invention may be used to express polypeptides of the invention. Host cells may be cultured under suitable conditions which
25 allow expression of the proteins of the invention. Expression of the polypeptides of the invention may be constitutive such that they are continually produced, or inducible, requiring a stimulus to initiate expression. In the case of inducible expression, protein

production can be initiated when required by, for example, addition of an inducer substance to the culture medium, for example dexamethasone or IPTG.

Polypeptides of the invention can be extracted from host cells by a variety of techniques known in the art, including enzymatic, chemical and/or osmotic lysis and
5 physical disruption.

Polypeptides of the invention may also be produced recombinantly in an *in vitro* cell-free system, such as the TnTTM (Promega) rabbit reticulocyte system.

ANTIBODIES

The invention also provides monoclonal or polyclonal antibodies to polypeptides of
10 the invention or fragments thereof. Thus, the present invention further provides a process for the production of monoclonal or polyclonal antibodies to polypeptides of the invention.

If polyclonal antibodies are desired, a selected mammal (e.g., mouse, rabbit, goat, horse, etc.) is immunised with an immunogenic polypeptide bearing an epitope(s) from a polypeptide of the invention. Serum from the immunised animal is collected and treated
15 according to known procedures. If serum containing polyclonal antibodies to an epitope from a polypeptide of the invention contains antibodies to other antigens, the polyclonal antibodies can be purified by immunoaffinity chromatography. Techniques for producing and processing polyclonal antisera are known in the art. In order that such antibodies may be made, the invention also provides polypeptides of the invention or fragments thereof
20 haptenised to another polypeptide for use as immunogens in animals or humans.

Monoclonal antibodies directed against epitopes in the polypeptides of the invention can also be readily produced by one skilled in the art. The general methodology for making monoclonal antibodies by hybridomas is well known. Immortal antibody-producing cell lines can be created by cell fusion, and also by other techniques such as
25 direct transformation of B lymphocytes with oncogenic DNA, or transfection with Epstein-Barr virus. Panels of monoclonal antibodies produced against epitopes in the

polypeptides of the invention can be screened for various properties; i.e., for isotype and epitope affinity.

An alternative technique involves screening phage display libraries where, for example the phage express scFv fragments on the surface of their coat with a large variety
5 of complementarity determining regions (CDRs). This technique is well known in the art.

Antibodies, both monoclonal and polyclonal, which are directed against epitopes from polypeptides of the invention are particularly useful in diagnosis, and those which are neutralising are useful in passive immunotherapy. Monoclonal antibodies, in particular, may be used to raise anti-idiotypic antibodies. Anti-idiotypic antibodies are
10 immunoglobulins which carry an "internal image" of the antigen of the agent against which protection is desired.

Techniques for raising anti-idiotypic antibodies are known in the art. These anti-idiotypic antibodies may also be useful in therapy.

For the purposes of this invention, the term "antibody", unless specified to the
15 contrary, includes fragments of whole antibodies which retain their binding activity for a target antigen. Such fragments include Fv, F(ab') and F(ab')₂ fragments, as well as single chain antibodies (scFv). Furthermore, the antibodies and fragments thereof may be humanised antibodies, for example as described in EP-A-239400.

Antibodies may be used in method of detecting polypeptides of the invention
20 present in biological samples by a method which comprises: (a) providing an antibody of the invention; (b) incubating a biological sample with said antibody under conditions which allow for the formation of an antibody-antigen complex; and (c) determining whether antibody-antigen complex comprising said antibody is formed.

Suitable samples include extracts tissues such as brain, breast, ovary, lung, colon,
25 pancreas, testes, liver, muscle and bone tissues or from neoplastic growths derived from such tissues.

Antibodies of the invention may be bound to a solid support and/or packaged into kits in a suitable container along with suitable reagents, controls, instructions and the like.

ASSAYS

The present invention provides assays that are suitable for identifying substances
5 which bind to polypeptides of the invention and which affect, for example, formation of the nuclear envelope, exit from the quiescent phase of the cell cycle (G0), G1 progression, chromosome decondensation, nuclear envelope breakdown, START, initiation of DNA replication, progression of DNA replication, termination of DNA replication, centrosome duplication, G2 progression, activation of mitotic or meiotic functions, chromosome
10 condensation, centrosome separation, microtubule nucleation, spindle formation and function, interactions with microtubule motor proteins, chromatid separation and segregation, inactivation of mitotic functions, formation of contractile ring, cytokinesis functions, chromatin binding, formation of replication complexes, replication licensing, phosphorylation or other secondary modification activity, proteolytic degradation,
15 microtubule binding, actin binding, septin binding, microtubule organising centre nucleation activity and binding to components of cell cycle signalling pathways.

In addition, assays suitable for identifying substances that interfere with binding of polypeptides of the invention, where appropriate, to components of cell division cycle machinery. This includes not only components such as microtubules but also signalling
20 components and regulatory components as indicated above. Such assays are typically *in vitro*. Assays are also provided that test the effects of candidate substances identified in preliminary *in vitro* assays on intact cells in whole cell assays. The assays described below, or any suitable assay as known in the art, may be used to identify these substances.

According to one aspect of the invention, therefore, we provide one or more
25 substances identified by any of the assays described below, viz, mitosis assays, meiotic assays, polypeptide binding assays, microtubule binding/polymerisation assays, microtubule purification and binding assays, microtubule organising centre (MTOC) nucleation activity assays, motor protein assay, assay for spindle assembly and function,

assays for dna replication, chromosome condensation assays, kinase assays, kinase inhibitor assays, and whole cell assays, each as described in further detail below.

CANDIDATE SUBSTANCES

A substance that inhibits cell cycle progression as a result of an interaction with a polypeptide of the invention may do so in several ways. For example, if the substance inhibits cell division, mitosis and/or meiosis, it may directly disrupt the binding of a polypeptide of the invention to a component of the spindle apparatus by, for example, binding to the polypeptide and masking or altering the site of interaction with the other component. A substance which inhibits DNA replication may do so by inhibiting the phosphorylation or de-phosphorylation of proteins involved in replication. For example, it is known that the kinase inhibitor 6-DMAP (6-dimethylaminopurine) prevents the initiation of replication (Blow, JJ, 1993, *J Cell Biol* 122,993-1002). Candidate substances of this type may conveniently be preliminarily screened by *in vitro* binding assays as, for example, described below and then tested, for example in a whole cell assay as described below. Examples of candidate substances include antibodies which recognise a polypeptide of the invention.

A substance which can bind directly to a polypeptide of the invention may also inhibit its function in cell cycle progression by altering its subcellular localisation and hence its ability to interact with its normal substrate. The substance may alter the subcellular localisation of the polypeptide by directly binding to it, or by indirectly disrupting the interaction of the polypeptide with another component. For example, it is known that interaction between the p68 and p180 subunits of DNA polymerase alpha-primase enzyme is necessary in order for p180 to translocate into the nucleus (Mizuno et al (1998) *Mol Cell Biol* 18,3552-62), and accordingly, a substance which disrupts the interaction between p68 and p180 will affect nuclear translocation and hence activity of the primase. A substance which affects mitosis may do so by preventing the polypeptide and components of the mitotic apparatus from coming into contact within the cell.

These substances may be tested using, for example the whole cells assays described below. Non-functional homologues of a polypeptide of the invention may also be tested

for inhibition of cell cycle progression since they may compete with the wild type protein for binding to components of the cell division cycle machinery whilst being incapable of the normal functions of the protein or block the function of the protein bound to the cell division cycle machinery. Such non-functional homologues may include naturally occurring mutants and modified sequences or fragments thereof.

Alternatively, instead of preventing the association of the components directly, the substance may suppress the biologically available amount of a polypeptide of the invention. This may be by inhibiting expression of the component, for example at the level of transcription, transcript stability, translation or post-translational stability. An example of such a substance would be antisense RNA or double-stranded interfering RNA sequences which suppresses the amount of mRNA biosynthesis.

Suitable candidate substances include peptides, especially of from about 5 to 30 or 10 to 25 amino acids in size, based on the sequence of the polypeptides described in the Examples, or variants of such peptides in which one or more residues have been substituted. Peptides from panels of peptides comprising random sequences or sequences which have been varied consistently to provide a maximally diverse panel of peptides may be used.

Suitable candidate substances also include antibody products (for example, monoclonal and polyclonal antibodies, single chain antibodies, chimeric antibodies and CDR-grafted antibodies) which are specific for a polypeptide of the invention. Furthermore, combinatorial libraries, peptide and peptide mimetics, defined chemical entities, oligonucleotides, and natural product libraries may be screened for activity as inhibitors of binding of a polypeptide of the invention to the cell division cycle machinery, for example mitotic/meiotic apparatus (such as microtubules). The candidate substances may be used in an initial screen in batches of, for example 10 substances per reaction, and the substances of those batches which show inhibition tested individually. Candidate substances which show activity in *in vitro* screens such as those described below can then be tested in whole cell systems, such as mammalian cells which will be exposed to the inhibitor and tested for inhibition of any of the stages of the cell cycle.

Polypeptide Binding Assays

One type of assay for identifying substances that bind to a polypeptide of the invention involves contacting a polypeptide of the invention, which is immobilised on a solid support, with a non-immobilised candidate substance determining whether and/or to what extent the polypeptide of the invention and candidate substance bind to each other. Alternatively, the candidate substance may be immobilised and the polypeptide of the invention non-immobilised.

In a preferred assay method, the polypeptide of the invention is immobilised on beads such as agarose beads. Typically this is achieved by expressing the component as a GST-fusion protein in bacteria, yeast or higher eukaryotic cell lines and purifying the GST-fusion protein from crude cell extracts using glutathione-agarose beads (Smith and Johnson, 1988). As a control, binding of the candidate substance, which is not a GST-fusion protein, to the immobilised polypeptide of the invention is determined in the absence of the polypeptide of the invention. The binding of the candidate substance to the immobilised polypeptide of the invention is then determined. This type of assay is known in the art as a GST pulldown assay. Again, the candidate substance may be immobilised and the polypeptide of the invention non-immobilised.

It is also possible to perform this type of assay using different affinity purification systems for immobilising one of the components, for example Ni-NTA agarose and histidine-tagged components.

Binding of the polypeptide of the invention to the candidate substance may be determined by a variety of methods well-known in the art. For example, the non-immobilised component may be labeled (with for example, a radioactive label, an epitope tag or an enzyme-antibody conjugate). Alternatively, binding may be determined by immunological detection techniques. For example, the reaction mixture can be Western blotted and the blot probed with an antibody that detects the non-immobilised component. ELISA techniques may also be used.

Candidate substances are typically added to a final concentration of from 1 to 1000 nmol/ml, more preferably from 1 to 100 nmol/ml. In the case of antibodies, the final concentration used is typically from 100 to 500 µg/ml, more preferably from 200 to 300 µg/ml.

5 ***Microtubule Binding/Polymerisation Assays***

In the case of polypeptides of the invention that bind to microtubules, another type of *in vitro* assay involves determining whether a candidate substance modulates binding of a polypeptide of the invention to microtubules. Such an assay typically comprises contacting a polypeptide of the invention with microtubules in the presence or absence of the candidate substance and determining if the candidate substance has an affect on the binding of the polypeptide of the invention to the microtubules. This assay can also be used in the absence of candidate substances to confirm that a polypeptide of the invention does indeed bind to microtubules. Microtubules may be prepared and assays conducted as follows:

15 ***Microtubule Purification and Binding Assays***

Microtubules are purified from 0-3h-old *Drosophila* embryos essentially as described previously (Saunders, *et al.*, 1997). About 3 ml of embryos are homogenized with a Dounce homogenizer in 2 volumes of ice-cold lysis buffer (0.1 M Pipes/NaOH, pH6.6, 5 mM EGTA, 1 mM MgSO₄, 0.9 M glycerol, 1 mM DTT, 1 mM PMSF, 1 µg/ml aprotinin, 1 µg/ml leupeptin and 1 µg/ml pepstatin). The microtubules are depolymerized by incubation on ice for 15 min, and the extract is then centrifuged at 16,000 g for 30 min at 4°C. The supernatant is recentrifuged at 135,000 g for 90 min at 4°C. Microtubules in this later supernatant are polymerized by addition of GTP to 1 mM and taxol to 20 µM and incubation at room temperature for 30 min. A 3 ml aliquot of the extract is layered on top of 3 ml 15% sucrose cushion prepared in lysis buffer. After centrifuging at 54,000g for 30 min at 20°C using a swing out rotor, the microtubule pellet is resuspended in lysis buffer.

Microtubule overlay assays are performed as previously described (Saunders *et al.*, 1997). 500 ng per lane of recombinant Asp, recombinant polypeptide, and bovine serum albumin (BSA, Sigma) are fractionated by 10% SDS-PAGE and blotted onto PVDF

membranes (Millipore). The membranes are preincubated in TBST (50mM Tris pH 7.5, 150 mM NaCl, 0.05% Tween 20) containing 5% low fat powdered milk (LFPM) for 1 h and then washed 3 times for 15 min in lysis buffer. The filters are then incubated for 30 minutes in lysis buffer containing either 1 mM GDP, 1 mM GTP, or 1 mM GTP- γ -S.

5 MAP-free bovine brain tubulin (Molecular Probes) is polymerised at a concentration of 2 μ g/ml in lysis buffer by addition of GTP to a final concentration of 1 mM and incubated at 37°C for 30 min. The nucleotide solutions are removed and the buffer containing polymerised microtubules added to the membranes for incubation for 1h at 37°C with addition of taxol at a final concentration of 10 μ M for the final 30 min. The blots are then

10 washed 3 times with TBST and the bound tubulin detected using standard Western blot procedures using anti- β -tubulin antibodies (Boehringer Mannheim) at 2.5 μ g/ml and the Super Signal detection system (Pierce).

It may be desirable in one embodiment of this type of assay to deplete the polypeptide of the invention from cell extracts used to produce polymerise microtubules.

15 This may, for example, be achieved by the use of suitable antibodies.

A simple extension to this type of assay would be to test the effects of purified polypeptide of the invention upon the ability of tubulin to polymerise *in vitro* (for example, as used by Andersen and Karsenti, 1997) in the presence or absence of a candidate substance (typically added at the concentrations described above). *Xenopus* cell-

20 free extracts may conveniently be used, for example as a source of tubulin.

Microtubule Organising Centre (MTOC) Nucleation Activity Assays

Candidate substances, for example those identified using the binding assays described above, may be screening using a microtubule organising centre nucleation activity assay to determine if they are capable of disrupting MTOCs as measured by, for

25 example, aster formation. This assay in its simplest form comprises adding the candidate substance to a cellular extract which in the absence of the candidate substance has microtubule organising centre nucleation activity resulting in formation of asters.

In a preferred embodiment, the assay system comprises (i) a polypeptide of the invention and (ii) components required for microtubule organising centre nucleation activity except for functional polypeptide of the invention, which is typically removed by immunodepletion (or by the use of extracts from mutant cells). The components
5 themselves are typically in two parts such that microtubule nucleation does not occur until the two parts are mixed. The polypeptide of the invention may be present in one of the two parts initially or added subsequently prior to mixing of the two parts.

Subsequently, the polypeptide of the invention and candidate substance are added to the component mix and microtubule nucleation from centrosomes measured, for
10 example by immunostaining for the polypeptide of the invention and visualising aster formation by immuno-fluorescence microscopy. The polypeptide of the invention may be preincubated with the candidate substance before addition to the component mix. Alternatively, both the polypeptide of the invention and the candidate substance may be added directly to the component mix, simultaneously or sequentially in either order.

15 The components required for microtubule organising centre formation typically include salt-stripped centrosomes prepared as described in Moritz *et al.*, 1998. Stripping centrosome preparations with 2 M KI removes the centrosome proteins CP60, CP190, CNN and γ -tubulin. Of these, neither CP60 nor CP190 appear to be required for microtubule nucleation. The other minimal components are typically provided as a
20 depleted cellular extract, or conveniently, as a cellular extract from cells with a non-functional variant of a polypeptide of the invention. Typically, labeled tubulin (usually β -tubulin) is also added to assist in visualising aster formation.

Alternatively, partially purified centrosomes that have not been salt-stripped may be used as part of the components. In this case, only tubulin, preferably labeled tubulin is
25 required to complete the component mix.

Candidate substances are typically added to a final concentration of from 1 to 1000 nmol/ml, more preferably from 1 to 100 nmol/ml. In the case of antibodies, the final

concentration used is typically from 100 to 500 $\mu\text{g/ml}$, more preferably from 200 to 300 $\mu\text{g/ml}$.

The degree of inhibition of aster formation by the candidate substance may be determined by measuring the number of normal asters per unit area for control untreated cell preparation and measuring the number of normal asters per unit area for cells treated with the candidate substance and comparing the result. Typically, a candidate substance is considered to be capable of disrupting MTOC integrity if the treated cell preparations have less than 50%, preferably less than 40, 30, 20 or 10% of the number of asters found in untreated cells preparations. It may also be desirable to stain cells for γ -tubulin to determine the maximum number of possible MTOCs present to allow normalisation between samples.

Motor Protein Assay

Polypeptides of the invention may interact with motor proteins such as the Eg5-like motor protein *in vitro*. The effects of candidate substances on such a process may be determined using assays wherein the motor protein is immobilised on coverslips. Rhodamine labeled microtubules are then added and their translocation can be followed by fluorescent microscopy. The effect of candidate substances may thus be determined by comparing the extent and/or rate of translocation in the presence and absence of the candidate substance. Generally, candidate substances known to bind to a polypeptide of the invention, would be tested in this assay. Alternatively, a high throughput assay may be used to identify modulators of motor proteins and the resulting identified substances tested for effects on a polypeptide of the invention as described above.

Typically this assay uses microtubules stabilised by taxol (e.g. Howard and Hyman 1993; Chandra and Endow, 1993 – both chapters in “Motility Assays for Motor Proteins” Ed Jon Scholey, pub Academic Press). If however, a polypeptide of the invention were to promote stable polymerisation of microtubules (see above) then these microtubules could be used directly in motility assays.

Simple protein-protein binding assays as described above, using a motor protein and a polypeptide of the invention may also be used to confirm that the polypeptide of the invention binds to the motor protein, typically prior to testing the effect of candidate substances on that interaction.

5 ***Assay for Spindle Assembly and Function***

A further assay to investigate the function of polypeptide of the invention and the effect of candidate substances on those functions is an assay which measures spindle assembly and function. Typically, such assays are performed using *Xenopus* cell free systems, where two types of spindle assembly are possible. In the "half spindle" assembly
10 pathway, a cytoplasmic extract of CSF arrested oocytes is mixed with sperm chromatin. The half spindles that form subsequently fuse together. A more physiological method is to induce CSF arrested extracts to enter interphase by addition of calcium, whereupon the DNA replicates and kinetochores form. Addition of fresh CSF arrested extract then induces mitosis with centrosome duplication and spindle formation (for discussion of
15 these systems see Tournabize and Heald, 1996).

Again, generally, candidate substances known to bind to a polypeptide of the invention, or non-functional polypeptide variants of the invention, would be tested in this assay. Alternatively, a high throughput assay may be used to identify modulators of spindle formation and function and the resulting identified substances tested for affects
20 binding of the polypeptide of the invention as described above.

Assays for DNA Replication

Another assay to investigate the function of polypeptide of the invention and the effect of candidate substances on those functions is as assay for replication of DNA. A number of cell free systems have been developed to assay DNA replication. These can be
25 used to assay the ability of a substance to prevent or inhibit DNA replication, by conducting the assay in the presence of the substance. Suitable cell-free assay systems include, for example the SV-40 assay (Li and Kelly, 1984, *Proc. Natl. Acad. Sci USA* 81, 6973-6977; Waga and Stillman, 1994, *Nature* 369, 207-212.). A *Drosophila* cell free replication system, for example as described by Crevel and Cotteril (1991), *EMBO J.* 10,

4361-4369, may also be used. A preferred assay is a cell free assay derived from *Xenopus* egg low speed supernatant extracts described in Blow and Laskey (1986, *Cell* 47,577-587) and Sheehan et al. (1988, *J. Cell Biol.* 106, 1-12), which measures the incorporation of nucleotides into a substrate consisting of *Xenopus* sperm DNA or HeLa nuclei. The nucleotides may be radiolabelled and incorporation assayed by scintillation counting. Alternatively and preferably, bromo-deoxy-uridine (BrdU) is used as a nucleotide substitute and replication activity measured by density substitution. The latter assay is able to distinguish genuine replication initiation events from incorporation as a result of DNA repair. The human cell-free replication assay reported by Krude, et al (1997), *Cell* 88, 109-19 may also be used to assay the effects of substances on the polypeptides of the invention.

Other In Vitro Assays

Other assays for identifying substances that bind to a polypeptide of the invention are also provided. For example, substances which affect chromosome condensation may be assayed using the *in vitro* cell free system derived from *Xenopus* eggs, as known in the art.

Substances which affect kinase activity or proteolysis activity are of interest. It is known, for example, that temporal control of ubiquitin-proteasome mediated protein degradation is critical for normal G1 and S phase progression (reviewed in Krek 1998, *Curr Opin Genet Dev* 8, 36-42). A number of E3 ubiquitin protein ligases, designated SCFs (Skp1-cullin-F-box protein ligase complexes), confer substrate specificity on ubiquitination reactions, while protein kinases phosphorylate substrates destined for destruction and convert them into preferred targets for ubiquitin modification catalyzed by SCFs. Furthermore, ubiquitin-mediated proteolysis due to the anaphase-promoting complex/cyclosome (APC/C) is essential for separation of sister chromatids during mitosis, and exit from mitosis (Listovsky et al., 2000, *Exp Cell Res* 255, 184-191).

Substances which inhibit or affect kinase activity may be identified by means of a kinase assay as known in the art, for example, by measuring incorporation of ^{32}P into a suitable peptide or other substrate in the presence of the candidate substance. Similarly,

substances which inhibit or affect proteolytic activity may be assayed by detecting increased or decreased cleavage of suitable polypeptide substrates.

Assays for these and other protein or polypeptide activities are known to those skilled in the art, and may suitably be used to identify substances which bind to a
5 polypeptide of the invention and affect its activity.

Whole Cell Assays

Candidate substances may also be tested on whole cells for their effect on cell cycle progression, including mitosis and/or meiosis. Preferably the candidate substances have been identified by the above-described *in vitro* methods. Alternatively, rapid
10 throughput screens for substances capable of inhibiting cell division, typically mitosis, may be used as a preliminary screen and then used in the *in vitro* assay described above to confirm that the affect is on a particular polypeptide of the invention.

The candidate substance, i.e. the test compound, may be administered to the cell in several ways. For example, it may be added directly to the cell culture medium or injected
15 into the cell. Alternatively, in the case of polypeptide candidate substances, the cell may be transfected with a nucleic acid construct which directs expression of the polypeptide in the cell. Preferably, the expression of the polypeptide is under the control of a regulatable promoter.

Typically, an assay to determine the effect of a candidate substance identified by
20 the method of the invention on a particular stage of the cell division cycle comprises administering the candidate substance to a cell and determining whether the substance inhibits that stage of the cell division cycle. Techniques for measuring progress through the cell cycle in a cell population are well known in the art. The extent of progress through the cell cycle in treated cells is compared with the extent of progress through the cell cycle
25 in an untreated control cell population to determine the degree of inhibition, if any. For example, an inhibitor of mitosis or meiosis may be assayed by measuring the proportion of cells in a population which are unable to undergo mitosis/meiosis and comparing this to the proportion of cells in an untreated population.

The concentration of candidate substances used will typically be such that the final concentration in the cells is similar to that described above for the *in vitro* assays.

A candidate substance is typically considered to be an inhibitor of a particular stage in the cell division cycle (for example, mitosis) if the proportion of cells undergoing that particular stage (i.e., mitosis) is reduced to below 50%, preferably below 40, 30, 20 or 10% of that observed in untreated control cell populations.

THERAPEUTIC USES

Many tumours are associated with defects in cell cycle progression, for example loss of normal cell cycle control. Tumour cells may therefore exhibit rapid and often aberrant mitosis. One therapeutic approach to treating cancer may therefore be to inhibit mitosis in rapidly dividing cells. Such an approach may also be used for therapy of any proliferative disease in general. Thus, since the polypeptides of the invention appear to be required for normal cell cycle progression, they represent targets for inhibition of their functions, particularly in tumour cells and other proliferative cells.

The term proliferative disorder is used herein in a broad sense to include any disorder that requires control of the cell cycle, for example, cardiovascular disorders such as restenosis and cardiomyopathy, auto-immune disorders such as glomerulonephritis and rheumatoid arthritis, dermatological disorders such as psoriasis, anti-inflammatory, anti-fungal, antiparasitic disorders such as malaria, emphysema and alopecia.

One possible approach is to express anti-sense constructs directed against polynucleotides of the invention, preferably selectively in tumour cells, to inhibit gene function and prevent the tumour cell from progressing through the cell cycle. Anti-sense constructs may also be used to inhibit gene function to prevent cell cycle progression in a proliferative cell. Another approach is to use non-functional variants of polypeptides of the invention that compete with the endogenous gene product for cellular components of cell cycle machinery, resulting in inhibition of function. Alternatively, compounds identified by the assays described above as binding to a polypeptide of the invention may

be administered to tumour or proliferative cells to prevent the function of that polypeptide. This may be performed, for example, by means of gene therapy or by direct administration of the compounds. Suitable antibodies of the invention may also be used as therapeutic agents.

5 Alternatively, double-stranded (ds) RNA is a powerful way of interfering with gene expression in a range of organisms that has recently been shown to be successful in mammals (Wianny and Zernicka-Goetz, 2000, Nat Cell Biol 2000, 2, 70-75). Double
10 stranded RNA corresponding to the sequence of a polynucleotide according to the invention can be introduced into or expressed in oocytes and cells of a candidate organism to interfere with cell division cycle progression.

 In addition, a number of the mutations described herein exhibit aberrant meiotic phenotypes. Aberrant meiosis is an important factor in infertility since mutations that affect only meiosis and not mitosis will lead to a viable organism but one that is unable to produce viable gametes and hence reproduce. Consequently, the elucidation of genes
15 involved in meiosis is an important step in diagnosing and preventing/treating fertility problems. Thus the polypeptides of the invention identified in mutant *Drosophila* having meiotic defects (as is clearly indicated in the Examples) may be used in methods of identifying substances that affect meiosis. In addition, these polypeptides, and
20 corresponding polynucleotides, may be used to study meiosis and identify possible mutations that are indicative of infertility. This will be of use in diagnosing infertility problems.

ADMINISTRATION

 Substances identified or identifiable by the assay methods of the invention may preferably be combined with various components to produce compositions of the
25 invention. Preferably the compositions are combined with a pharmaceutically acceptable carrier or diluent to produce a pharmaceutical composition (which may be for human or animal use). Suitable carriers and diluents include isotonic saline solutions, for example phosphate-buffered saline. The composition of the invention may be administered by

direct injection. The composition may be formulated for parenteral, intramuscular, intravenous, subcutaneous, intraocular or transdermal administration. Typically, each protein may be administered at a dose of from 0.01 to 30 mg/kg body weight, preferably from 0.1 to 10 mg/kg, more preferably from 0.1 to 1 mg/kg body weight.

5 Polynucleotides/vectors encoding polypeptide components (or antisense constructs) for use in inhibiting cell cycle progression, for example, inhibiting mitosis or meiosis, may be administered directly as a naked nucleic acid construct. They may further comprise flanking sequences homologous to the host cell genome. When the polynucleotides/vectors are administered as a naked nucleic acid, the amount of nucleic
10 acid administered may typically be in the range of from 1 µg to 10 mg, preferably from 100 µg to 1 mg. It is particularly preferred to use polynucleotides/ vectors that target specifically tumour or proliferative cells, for example by virtue of suitable regulatory constructs or by the use of targeted viral vectors.

Uptake of naked nucleic acid constructs by mammalian cells is enhanced by
15 several known transfection techniques for example those including the use of transfection agents. Example of these agents include cationic agents (for example calcium phosphate and DEAE-dextran) and lipofectants (for example lipofectamTM and transfectamTM). Typically, nucleic acid constructs are mixed with the transfection agent to produce a composition.

20 Preferably the polynucleotide, polypeptide, compound or vector described here may be conjugated, joined, linked, fused, or otherwise associated with a membrane translocation sequence.

Preferably, the polynucleotide, polypeptide, compound or vector, etc described here may be delivered into cells by being conjugated with, joined to, linked to, fused to, or
25 otherwise associated with a protein capable of crossing the plasma membrane and/or the nuclear membrane (i.e., a membrane translocation sequence). Preferably, the substance of interest is fused or conjugated to a domain or sequence from such a protein responsible for the translocational activity. Translocation domains and sequences for example include

domains and sequences from the HIV-1-trans-activating protein (Tat), *Drosophila* Antennapedia homeodomain protein and the herpes simplex-1 virus VP22 protein. In a highly preferred embodiment, the substance of interest is conjugated with penetratin protein or a fragment of this. Penetratin comprises the sequence

- 5 RQIKIWFQNRRMKWKK and is described in Derossi, *et al.*, (1994), *J. Biol. Chem.* 269, 10444-50; use of penetratin-drug conjugates for intracellular delivery is described in WO/00/01417. Truncated and modified forms of penetratin may also be used, as described in WO/00/29427.

- 10 Preferably the polynucleotide, polypeptide, compound or vector according to the invention is combined with a pharmaceutically acceptable carrier or diluent to produce a pharmaceutical composition. Suitable carriers and diluents include isotonic saline solutions, for example phosphate-buffered saline. The composition may be formulated for parenteral, intramuscular, intravenous, subcutaneous, intraocular or transdermal administration.

- 15 The routes of administration and dosages described are intended only as a guide since a skilled practitioner will be able to determine readily the optimum route of administration and dosage for any particular patient and condition.

- 20 The invention will now be further described by way of Examples, which are meant to serve to assist one of ordinary skill in the art in carrying out the invention and are not intended in any way to limit the scope of the invention.

EXAMPLES

Generation and Identification of Lethal, Semi-Lethal and Sterile Third Chromosome Mutants Having Defects in Mitosis and/Or Meiosis, and Second Chromosome Mutants Having Defects in Imaginal Disc Development By P-Element

5 Insertion Mutagenesis

P-element mutagenesis

Transposable elements are widely used for mutagenesis in *Drosophila melanogaster* as they couple the advantages of providing effective genetic lesions with ease of detecting disrupted genes for the purpose of molecular cloning. To achieve near
10 saturation of the genome with mutations resulting from mobilisation of the P-lacW transposon (a P-element marked with a mini-white gene, bearing the *E.coli lacZ* gene as an enhancer trap, and an *E.coli* replicon and ampicillin resistance gene to facilitate 'plasmid rescue' of sequences at the site of the P-insertion), *Drosophila* females that are homozygous for *P-lacW* (inserted on the X chromosome) are crossed with males carrying
15 the transposase source P($\Delta 2-3$) (Deak et al., 1997). Random transpositions of the mutator element are then 'captured' in lines lacking transposase activity. Stable, or balanced, stocks bearing single lethal *P-lacW* insertions are made.

More than 41,000 lines are derived, of which approximately one-half are on the third chromosome. Originally some 3100 lethal or strong semi-lethal lines (in homozygous
20 conditions) are identified. During preliminary characterisation unstable lines and clusters of the same mutation event are eliminated leaving 2460 lines to be characterised.

Screening for Mitotic and Meiotic Defects

About half of the mutants in the collection are embryonic lethals. We have carried out cytological screens of the 1155 lines that comprise late larval lethals, pupal lethals,
25 pharate and adult semi-lethals for defective mitosis in the developing larval CNS. This has identified 69 mutations falling into 43 complementation groups that affect all stages of the mitotic cycle. The cytological screens involve examining orcein-stained squashed preparations of the larval CNS to detect abnormal mitotic cells. In lines where defects are

identified, the larval CNS is subjected to immunostaining to identify centromeres, spindle microtubules and DNA for further examination. This leads to clarification of the mitotic defect.

As a set of common functions are essential to both mitosis and meiosis, we then
5 identify mutations resulting in sterility and failed progression through male meiosis. This involves examining squashed preparations larval, pupal or adult testes by phase contrast microscopy. We examine "onion stage" spermatids in the 519 pupal and pharate lethal lines and 463 adult "semi-lethal" and viable lines for variations in size and number of nuclei which provides an indication of whether there have been defects in either
10 chromosome segregation or cytokinesis, respectively. A total of 54 lines of the 519 pupal and pharate lethal lines and 22 of the adult lines show such defects. However, another 67 lines show male sterility without having onion-stage defects. 12 lines showing onion stage defects have been scored as having mitotic defects in the independent cytological screen of squashed preparations of the larval CNS. Twelve further lines with onion stage defects
15 show female sterility and of these, 10 show maternal effect mitotic defects in syncytial embryos. Thus greater than one half of the meiotic mutants scored appear to represent cell division functions specific to male meiosis or have targeted male germ-line specific enhancer elements, thus revealing their meiotic function but in this test not their mitotic function.

20 Further characterisation of testis preparations of each line by phase-contrast microscopy with and without staining with Hoechst to reveal DNA defined 6 broad categories of meiotic mutants:

8 mutants from the collection show defects in meiotic entry or at early stages in the first meiotic division (MF1-8).

25 18 mutants (15 complementation groups) show abnormal meiotic spindles (AB1-16). Mutants in this group almost invariably show an associated weak defect in cytokinesis, and 7 show a strong defect in spermatid differentiation. 3 of these mutants

also show mitotic defects in larval brains or in embryos derived from homozygous mutant mothers.

18 mutants (16 complementation groups) also show abnormal meiotic spindles that are strongly multipolar (MUL1-15). Three of these also show maternal effect mitotic abnormalities of multipolar spindles in syncytial embryos.

4 mutants (3 complementation groups) show strong defects at all stages of spermatogenesis from the pre-meiotic stages to spermatid elongation stages (PL1-3). In this respect they resemble the *polo*¹ mutation.

4 mutants show segregation defects as indicated by spermatid nuclei of heterogeneous sizes (SEG1-4). The spindles appear normal but all have what are either chromosome bridges or lagging chromosomes. One of these also shows a maternal effect.

9 mutants (7 complementation groups) show predominant cytokinesis defects. Two complementation groups also have cytokinesis defects in mitotic cells in the larval brain.

In the Examples below, the designations MF, AB, MUL, PL, SEG or CK are included in the category description where available. Further phenotype information for each mutant described in the results section is provided in the "Phenotype" field. There is considerable overlap between these categories, and it will be of much interest to distinguish between mutants in which the primary defect results in secondary consequences, and mutants that affect more than one aspect of spermatogenesis, as for example appears to be the case with *polo* mutants (Sunkel and Glover, 1988; Carmena et al, 1998).

In the Examples, lines exhibiting mitotic and meiotic phenotypes are categorised generally into four categories:

Category 1 : Failure to complete cytokinesis

Category 2 : Failure to enter M-phase

Category 3: Metaphase arrest

Category 4: Anaphase defect

Category 5: Small Imaginal Discs (Block to Proliferation; see below)

5 Category 1 phenotypes are exhibited by mutations in Examples 1 to 14; while Category 2 phenotypes are exhibited by mutations in Examples 15 to 19. Category 3 phenotypes are exhibited by mutations in Examples 20 to 30, Category 4 phenotypes are exhibited by mutations in Examples 31 to 53. Mutations in Examples 54 to 74 exhibit a Category 5 phenotype.

10 ***Generation and identification of second chromosome mutants having small or no imaginal discs.***

In the case of the second chromosome the flies used were from a second chromosome P-element collection established in Szeged, Hungary (Torok et al., 1993). The process of P-element insertion mutagenesis is essentially as described above. 15475
15 insertions were recovered, of which 2711 were lethal or semi-lethal. After elimination of clusters of identical mutants, 2399 lines representing 1748 independent lethal insertions were recovered. Lines were chosen from the second chromosome collection on the basis of having small or no imaginal discs, to indicate a disruption in cell cycle progression that leads to underdevelopment of the discs. All the second chromosome mutants referred to in
20 the results section are noted under the "Phenotype" field as "second chromosome, small imaginal discs" and comprise Category 5.

Cytological Mapping of the P-Element Insertion Sites

The site of insertion of the P-element in each mutant line was determined by *in situ* hybridisation of P-element DNA to salivary gland polytene chromosomes as described in
25 Saunders et al., 1989. Wandering third stage larvae were dissected and fixed as described and incubated with biotin-labeled DNA made from the *P-lacW* plasmid. After signal

detection chromosomes were stained with Giemsa and examined by microscopy and signals indicating the presence of P elements were assigned to polytene chromosome bands referring to the Bridges map (Lefevre, 1976). In the majority of cases a single P element was detected, only 10% of lines having multiple (two or three) insertions. The site of insertion is given as the "Map Position" field in the results section (for example 77B)

Plasmid Rescue of P-Elements from Mutant Drosophila Lines

Genomic DNA was isolated from adult flies by the method of Jowett et al., 1986, and plasmid rescue from the genomic DNA was performed according to Pirrotta et al., 1986. This allows the recovery of genomic DNA adjacent to the P-element which facilitates the identification of the site of P-element insertion and of genes which may be disrupted by the insertion. Essentially, genomic DNA derived from about 200 flies was digested with a restriction enzyme known to have a site within the P-element (EcoRI or SacII for cloning sequences to the left of the element, or XbaI, BglII, PstI or BamHI for sequences to the right of the element). The digested DNA was ligated overnight, and plasmids recovered by electroporation of the ligated DNA into *E. coli* XL1-blue competent cells. Appropriate primers from within the P-lacW sequence were used to determine the sequence of the genomic DNA flanking the element (on average, 400 bp of sequence were obtained). The rescue sequences are provided in the results section under the heading "Rescue sequence". Where more than one sequence was recovered, the orientation of each sequence is also given.

Sequence Analysis of P Element Insertion Lines

Sequences flanking the insertion site of the P-element were derived by P element rescue as described above. In some cases sequence was obtained from only one side of the insertion, while in other cases sequences were obtained from both sides of the insertion.

As a first step, each P element rescue sequence was used to search a total database of *Drosophila melanogaster* sequences (database of the Berkley *Drosophila* Genome project) using the BLASTN program (which compares a nucleic acid sequence with a nucleic acid database, (Altschul and Lipman 1990)) with default parameters.

The search may identify a number of different types of match including *Drosophila* ESTs, known *Drosophila* genes and cloned genomic regions.

The ability to identify genes already known to be essential for cell cycle progression using this approach was confirmed, in this example, by the rescue sequence
5 obtained from line 1324/8 which mapped to the 77B locus which was used to search the database. A BLASTN search identified a number of matching *Drosophila* ESTs, a match with the known cell cycle regulatory gene *polo* and a cloned genomic region designated CSC: AC018188. These matches are recorded in the results sections under the field headings "*Drosophila* ESTs", "*Drosophila* gene hit" and "Genomic hit, Accession No.",
10 respectively. Any entries under "*Drosophila* gene hit" are further annotated with "(BLASTN with Rescue sequence)" to show that the match was obtained using the rescue sequence rather than a *Drosophila* EST or genomic clone ORF (see below). Accession numbers of ESTs, genes and genomic clones are provided where known. Genomic clones designations often include the Genbank designation as part of a longer designation.
15 However the Genbank designation is always the code beginning with "AC" and followed by six digits.

Where an EST was identified, this was subsequently used to search using the BLASTX program (default parameters) against databases of sequences from *Drosophila* and Homo sapiens (databases of the National centre for Biotechnology Information
20 (NCBI), National Library of Medicine, National Institute of Health, USA). In the case of line 1104/16, the search identified a known human gene, phosphatidylinositol transfer protein (accession no. P48739) implying a novel function for this protein in cytokinesis. Human Homologues identified as a result of a BLASTX search using a *Drosophila* EST are shown in the results section under the heading "Human homologues" and annotated
25 with "(BLASTX with EST)". *Drosophila* genes identified as a result of a BLASTX search using a *Drosophila* EST are shown in the results section under the heading "*Drosophila* gene hit" and annotated with "(BLASTX with EST)".

Where no *Drosophila* gene was identified using the initial BLASTN search but a matching genomic clone was found (a Bac or P1 clone often in excess of 100 kilobases), a

20 kilobase segment of this genomic sequence (10 kilobases either side flanking the site of the P-element insertion) was subjected to a number of analyses.

If the rescue sequence matched sequences that lie within a known gene present within the genomic clone then these are presented under the heading "*Drosophila* gene hit
5 (BLASTN with Rescue sequence". The known gene sequence was then used in a BLASTX search of a human database (NCBI – see above) to identify any human homologues. These are shown in the "Human homologue" field and annotated with "(BLASTX with *Drosophila* gene)".

If the rescue sequence does not match any sequences that lie with a known gene
10 within the genomic clone then the occurrence of ORFs within the 20 kilobase genomic segment was predicted using the Genscan programme (Burge and Karlin, 1997). Where the P-element was observed to be inserted into the coding region or within the 5' untranslated region (which we defined as within 2 kilobases of the predicted start of the coding region) we assume the P element to be capable of disrupting the expression of the
15 predicted gene. Each predicted open reading frame (or predicted coding sequence) was then used to search *Drosophila* and human databases using the TBLASTN program (compares a protein query sequence against a nucleotide sequence database dynamically translated in all reading frames) and/or the TBLASTX program (compares a nucleotide query sequence dynamically translated in all reading frames against a nucleotide sequence
20 database dynamically translated in all reading frames) to determine whether the predicted open reading frame corresponded to a known gene. Typically, TBLASTX is only used when no matches are found using TBLASTN.

Where the TBLASTN search found a known *Drosophila* gene, then this is indicated in the results in the "*Drosophila* gene hit" field, annotated with "(TBLASTN
25 with predicted ORF)". The *Drosophila* gene sequence was then typically used to search a human database (NCBI – see above) to identify any human homologues using BLASTX. These are shown in the "Human homologue" field and annotated with "(BLASTX with *Drosophila* gene)".

Where the TBLASTN and/or TBLASTX search found a known human gene, then this is indicated in the results in the "Human homologue" field, annotated with "(TBLASTN (or TBLASTX) with predicted ORF)".

5 If the TBLASTN and/or TBLASTX search found no *Drosophila* or human genes, then it was assumed that the original ORF corresponds to a novel gene. If the TBLASTN search found no *Drosophila* genes but identified a human homologue, then it was assumed that the original ORF corresponds to a novel *Drosophila* homologue of a known human gene.

10 ***Additional Sequence Analysis using the Annotated D. melanogaster Sequence (GadFly).***

Rescue sequences were also used to search the fully annotated version of the *Drosophila* genome (GadFly; Adams, et al., 2000, Science 287, 2185-2195), using GlyBLAST at the Berkeley *Drosophila* Genome Projects web site to identify the genome segment (usually approximately 200-250 kb) containing the P-element insertion site. The
15 graphic representation of the genomic fragment available at GadFly allows the identification of all real and theoretical genes which flank the site of insertion. Candidate genes where the P-element is either inserted within the gene or close to the 5' end of the gene were identified. In GadFly, the *Drosophila* genes are given the designation CG (Complete gene) and usually details of human homologues are also given. In most cases,
20 this data confirms the data derived from the sequence analysis procedure described above, and in some cases new data is obtained. Where available both sets of data are included in the individual Examples described below. To identify further candidate human homologues, BLASTP (amino acid query sequence against amino acid database) searches with *Drosophila* sequences are used against the human genome project database and also
25 the Ensembl dataset. The Ensembl dataset comprises GeneWise gene predictions using a protein template where possible or Genscan followed by BLAST confirmation via protein, cDNA or EST hits. These are matched using WUBLASTP with default parameters (Altschul et al., 1990, *J Mol Biol* 215, 403-10). The results are filtered to contain only potential homologues. Only matches with the identity of more than 50% and length of
30 more than 50 amino acids are included.

Confirmation of Cell Cycle Involvement of Candidate Genes Using Double Stranded RNA Interference (RNAi)

P-elements usually insert into the region 5' to a *Drosophila* gene. This means that there is sometimes more than one candidate gene affected, as the P-element can insert into the 5' regions of two diverging genes (one on each DNA strand). In order to confirm which of the candidate genes is responsible for the cell cycle phenotype observed in the fly line, we use the technique of double stranded RNA interference to specifically knock out gene expression in *Drosophila* cells in tissue culture (Clemens, et al., 2000, *Proc. Natl. Acad. Sci. USA*, 6499-6503). The overall strategy is to prepare double stranded RNA (dsRNA) specific to each gene of interest and to transfect this into Schneider's *Drosophila* line 2 to inhibit the expression of the particular gene. The dsRNA is prepared from a double stranded, gene specific PCR product with a T7 RNA polymerase binding site at each end. The PCR primers consist of 25-30 bases of gene specific sequence fused to a T7 polymerase binding site (TAATACGACTCACTATAGGGACA), and are designed to amplify a DNA fragment of around 500bp. Although this is the optimal size, the sequences in fact range from 450 bp to 650 bp. Where possible, PCR amplification is performed using genomic DNA purified from Schneider's *Drosophila* line 2 as a template. This is only feasible where the gene has an exon of 450 bp or more. In instances where the gene possesses only short exons of less than 450 bp, primers are designed in different exons and PCR amplification is performed using cDNA derived from Schneider's *Drosophila* line 2 as a template.

A sample of PCR product is analysed by horizontal gel electrophoresis and the DNA purified using a Qiagen QiaQuick PCR purification kit. 1µg of DNA is used as the template in the preparation of gene specific single stranded RNA using the Ambion T7 Megascript kit. Single stranded RNA is produced from both strands of the template and is purified and immediately annealed by heating to 90 degrees C for 15 mins followed by gradual cooling to room temperature overnight. A sample of the dsRNA is analysed by horizontal gel electrophoresis.

3µg of dsRNA is transfected into Schneider's *Drosophila* line 2 using the transfection agent, Transfect (Gibco) and the cells incubated for 72 hours prior to fixation.

The DNA content of the cells is analysed by staining with propidium iodide and standard FACS analysis for DNA content. The cells in G1 and G2/S phases of the cell cycle are visualised as two separate population peaks in normal cycling S2 cells. In each experiment, Red Fluorescent Protein dsRNA is used as a negative control. In some cases
 5 the phenotype is confirmed by fixing cells on poly-lysine covered slides which are then stained for DNA using DAPI and for tubulin using an anti-tubulin antibody YL1/2 and appropriate fluorescent secondary antibody to visualise aberrant mitoses.

It should be noted that RNAi could not confirm phenotype in all cases. This is to be expected as the method relies on the ability of dsRNA to prevent new protein
 10 expression. Consequently, it is necessary that S2 cells express the specific cDNA of the gene in question, and also that the protein is turned over rapidly. It would therefore probably be difficult to sufficiently reduce levels of very stable proteins using this approach.

The layout of a typical entry in the results section is shown below. Not all fields
 15 present in the actual results section contain information for each individual *Drosophila* line described.

TYPICAL RESULTS LAYOUT

20	Line ID	- <i>Drosophila</i> line designation
	Category	- Description of phenotype
	Reversion	- R = revertant, NR = non revertant, ? = not determined
	Map Position	- according to the Bridges map (Lefevre, 1976).

25	Rescue ID
	Rescue Sequence
	[nucleotide sequence]

Genomic hit, Accession No.

30	Associated ORF
	GENSCAN_predicted_peptide [results of Genscan - amino acid sequence]
	GENSCAN_predicted_CDS [results of Genscan nucleotide sequence]

35	<i>Drosophila</i> Gene Hit
	(BLASTN with rescue sequence)

(TBLASTN (or TBLASTX) with predicted ORF)
(BLASTX with EST)

Human Homologue

- 5 (BLASTX with *Drosophila* gene)
(TBLASTN (or TBLASTX) with predicted ORF)
(BLASTX with EST)
Drosophila EST

- 10 **Annotated *Drosophila* genome genomic segment**
Annotated *Drosophila* genome Complete gene candidate
Human homologue of Complete gene candidate

Putative function Derived from homologies or *Drosophila* experimental data

- 15 **Confirmation by RNAi** Description of Facs analysis DNA content profile

A specific example is as follows:

20

Line ID 1324/8
Category Mitotic defects in brain: metaphase arrest
(overcondensation, some circular chromosomes, no anaphases,
very high mitotic index, metaphase (or less aligned) with bipolar
spindle, no CP190 staining)
Reversion R
Map Position 77B

25

Rescue ID B1E

30

Rescue Sequence

GTTTTGCCCATCGATTGCACGAAAACCAAGCACAAAGCGGAGAACGCGCCGA
AACCGTTCGATTTTTTAAATGCCAAATGAATTGGACGTGAAGCGTCAGCTGA
ATTGGTGTGCCCGTTTCGGTGGCTATCGCACACTTTCTGGTATTTATCGCGGTA
TTTTGTTGAGTGTTGAACAACAAATTCTATGGCCGTTACCCTTTGAATTTACT
35 TACTGGCGTTTACTCTGTTTCAATTGAGCGCAATATTTTTTCCTATTGCTCTGC
GCAAACTGTGTTTTAACCGCTATTTATTTGAAAATCTACAAAACTAACCGTT
TACATTTTTGAAATTTCCAAAAGGGTTTTCCATAAATTGAGTTTTACTAAAACC
AGTCCAACGGTCCAACCTTATATTGTTAGAAGCCCCTTTTCCTAATTTGAATTG
GCTTGCAAACGTTTTCTGAATTTAAAAATACTGCCACCCTTGTTAATTGCAGG
40 TTTTCCGAATCCCTGATTTGTTGTTTTAAAAAGAAAATTTATTAGAAACAGCTA
TCTCAACC

Genomic hit, Accession No. CSC:AC018188

***Drosophila* Gene Hit** Polo (X63361)

45

Human Homologue BLASTX PLK-1 (P53350)

***Drosophila* EST** several including LD11851 (AA392613) which match polo

Annotated *Drosophila* genome genomic segment AE003514

Annotated *Drosophila* genome Complete gene candidate CG12306

Human homolog of Complete gene candidate

1e-169 1709658 P53350

PLK1_HUMAN

SERINE/THREONINE-
PROTEIN KINASE PLK
(PLK-1)

5

Putative function Serine/threonine kinase known to be required for mitosis

- 10 **Confirmation by RNAi** Reduced G1 and G2/M peaks indicating fewer cycling cells, microscopy analysis of DNA and tubulin staining identified monopolar spindles characteristic of polo mutation in *Drosophila*.

CATEGORY 1: FAILURE TO COMPLETE CYTOKINESIS**Example 1 (Category 1)**

Line ID 1031/14
5 Category Mitotic defects in brain: cytokinesis defect (polyploidy)
Reversion R
Map Position 74B

10 Rescue ID 2A3B
Rescue Sequence 1
CCCCGGAACATATGTTTCAGTGTGGCCGCAGCAGAGTTGTCAAAACACGCTCCC
CAATGAAATAACCTAAATGTGCCATCACTGTTACTTAAACAGTTTCTGTTACTTT
TCTAGCGGCATGTCAAAAAAACAAAAATATAGAAAATGCTAAATATATATTG
15 GACTAATGTGTTTAAATGTAACCTTACACTAGTAACAGATCCCCATTAATAAAA
GCCAAACTCTAAAATTCTGCCACAAGTACTATTTCTCACGTAACACCTTACTA
ACGGATTTTCACATGATATCTACGACAAGAACTGTTTGCTGATATAAAATTGC
TATCACCGCTTTCCGTAAACACTTTTACACTGATGGATTACAAGTTCAATTAAT
ACATCAACTTACCTTAACAATTTTAAAGACAATAACACTCCCACAATTTAATT
20 CAACCTACACCGCTTGATAATCAGCTGTTCTGTACAAAAACAATAACACTGT
TAACAACAGCGCACAGTGGATAATACAGTCCTAAAGGCAATATACCCATTG
GCATTTTT

Rescue ID 2A3S
25 Rescue Sequence 2
TTCCGGGGAGAATGGCTGCGATTTTCGCGTCGGTAAAAATAGCAAATACTCGTTA
ATGTGCTGTGGGAACGCTTCCTCCCCGGCCCCAAAGTGGCCCCGAAGAAAGTGA
GCAAAATGTGCGCGCCGCAAGATAGTCGCCGCGAACAACGATAGTGACGAAA
GTGATTTAATTCAACTACCAGCACTCCCGCAAATACGATGAGTATGTCGCGCGG
30 CGGCAACACAACTCTGGACTTGCGAGCCGCTCCTGGCGGAGAGCGATGTCGGAA
ACAGGGAGCTGGAGGAGAAGATGGGCGGATCGGCGGATCGGTCATCGCTGCTC
GATGGATCCGGTTTCGAAGGAGCTGAGTCACCGGGAACGCGAGGACTCGGCGTT
GTTTCGTCAAGAAGATCGGGAGCGCCTTGTTCTATGGCTTGTCCTCCTTCATGATT
ACGGTGGTAAACAAGACGGTGCTTACCTCCTACCACTTCCCCTCGTTCCTGTTCC
35 TCAGCCTCGGGCAACTTACTGCTAGCATTGTGGTCCTGGGCATGGGCAAAGCGC
CTGAAAATGGTGAACTTTTCCCCTTTTGCAGAGGAATACCTTCGCCAAGATCTTT
CCGCTGCCACTGATATTTCTGGGAAACATGATGTTTGGACTGGGTGGCACAAAA
ACCTTGAGTCTGCCCATGTTTCGCAGCCCTACGAC

40 Genomic hit, Accession No. AC019515

Associated ORF

Genscan ORF1 predicted sequences:>15:31:57|GENSCAN_predicted_peptide_4|373_aa

MSMSRGGNTTLDLQPLLAESDVGNRELEEKMGGSADRSSLLDGS GSKELSHRER
 EDSALFVKKIGSALFYGLSSFMITVNVKTVLTSYHFPSFLFLSLGQLTASIVVLGMG
 KRLKLVNFPPLQRNTFAKIFPLPLIFLGNMMFGLGGTKTSLPMFAALRRFSILMT
 MLELKILGLRPSNAVQVSVMYAMIGGALLAASDDL SFNMRGYTYVMITNALTASN
 5 GVVYVKKKLDTSEIGKYGLMYNSLFMFLPALALNYVTGNLDQALNFEQWNSV
 FVVQFLLSCVMGFILSYSTILCTQFNSALTTTIVGCLKNICVTYLGFMFIGGDYVFSW
 LNCIGINISVLASLLYTYVTFRRKRAPDKQDHL PSTRGENV

>15:31:57|GENSCAN_predicted_CDS_4|1122_bp

10 atgagtatgtcgcggcggcgaacacaactctggactgcagccgctcctggcggagagcgtatcggaaacaggagctgga
 ggagaagatggcggatcggcggatcggatcgcctgctcgtatggatccgggtcgaaggagctgagtcaccgggaacgcgag
 gactcggcgttgctcgtcaagaagatcgggagcgcctgttctatggctgtcctcctcatgattacgggtgtaacaagacgggtgc
 ttacctcctaccacttcccctcgttcctgttctcagcctcgggcaacttactgctagcattgtggtcctgggcatgggcaagcgct
 gaaattggtgaactttccccctcgtcagaggaataccttcgccaatctttcgcctgactgatattctgggaacatgatgtttg
 15 gactgggtggcacaaaaaccttgagtctgccatgttcgcagccctacgacgcttctctatcctgatgacctgctgctggagctca
 agatcctgggactgcgacctcgaatgcgggtcaggtcagcgtatagcgaatgatcgggtggagcgtgctgcccgcctctgatga
 tctgtccttaacatgaggggtacatctatgtgatgactaaccgcttgaccgcctcgaatggcgatatgtgaagaaaaaac
 gacacctcgagatcggaaagtacggcctaattactacaactcgtgtttatgtttctgctgccctggccctcaactatgttacag
 ggaatctagatcaggcgtgaactttgaacaatggaatgactcagtggttggtgcagttcctgctcagttgcgttatgggttcac
 20 ctatcgtacagcaccatcctgtgcacgaattcaactcggcgtgaccaccaccattgtgggatgcctgaaaaacatctgcgtaac
 atatctgggcattgctgaggcgactacgtctctcgtggcgaactgtattggatcaacatcagcgtgctgtagtctgctct
 acacgtacgtcacttttcggcggagcgggtcccgcataagcaggaccacttgccagcaccgcggcgagaatgtctag

25 **Human Homologue** (TBLASTN with ORF1): KIAA0260 gene (D87449) and putative
 Sqv-7-like protein (AJ005866)
Drosophila EST CK00510 (AA140776)

Annotated Drosophila genome genomic segment AE003524
 30 **Annotated Drosophila genome Complete gene candidate** CG3874 – novel glucose-6-
 phosphate transporter

Human homologue of Complete gene candidate EMBL:D87449 protein
 KIAA0260_id:BAA13390
 35 gi:166578 Similar to a
 C.elegans protein encoded in
 cosmid C52E12 (U50135) and
 Ensembl predicted gene
 ENSG00000024527
 Clone:AL133320
 40 Contig:AL133320.00001
 8.10E-95

Putative function Sugar modification protein similar to proteins involved in
 Drosophila cytokinesis and signalling

45 **Confirmation by RNAi** Marked increased G1 and S peak indicating mainly arrest in
 G1

Example 2 (Category 1)

Line ID 1066/5
Category Male semi-sterile, Meiotic defects in testis: cytokinesis defects, segregation defects.
 5 (Seg-01/62)
Reversion ?
Map Position 89B
 10 **Rescue ID** F9E
Rescue Sequence
 GTATACCATAGAGAATATGATGAAGAAGGACTGTAAGAAGATCCTTCAGTG
 AATTTGACTGCTGACGTCGATCGGAACCTTGCTGCGCTGACGTACAAAATCGCG
 AAGTGAATAAATAATATGGATGAGACCCTGTTTCGCCGACATATACAATAGTG
 15 CTCAAGACCTAATGGAATTATACGTTAATAACCAGCCACATTTCTTAGATATTT
 CTAATATGAGCCATCTGCTGCAGGTTCTTTCCAATATCTAATTCTAGATCTTCT
 TCGAATACGACCTTTTTGGCCATGAAACGATGATTTGCCACTTCATTACAAG
 CATTAAATTTGTCATGATTCTCTTAAGCGTGCACCTTTATCTGAAAGTCTGAACAG
 CTGGCTGCGAAATGGATCCCCGGGATTGGAGATGGCAAAGTAAATCTGTCTCTCG
 20 CTACAAACAAGTGGGCACCACTGGGCATTCTGGGGAATAGGGATATGGGTTGG
 GAATGGGGATATATTGTGGCATTGGCGAAAGGTCGCTATGC

Genomic hit, Accession No. CSC:AC019750

25 **Associated ORF**
 >16:04:57|GENSCAN_predicted_peptide_4|418_aa
 MKPIPNESKGTAAVGDATVVHDTVCTLFAVELDPYLRSSMGMRTTRAQSGALLL
 QLLAVADGGFAAHICACKCRLRLPHVTCCNRNPFKATAKAKGQAVSSTKPNQL
 CFHGCCGWITTKGETFTENSPSIMSGFAWERHSLGECVVVAGTEQILLIGRTLIGR
 30 MSHTQTDSTSPFVVDCHSQLCGSKCKCICVSVGFVVRPSCQRFDMKIVWANLAM
 QKRFLGAAIADMCCRNSVTWCKLQLDPVKPIDERADGSGGLALVTKVCDNNIV
 HYVVVAGVTGSQSRSLQPLRSGQNESTEQWPRTKGEGGFNNNSRNKHSAPT
 QEQQELWQKQLQDQRDDCHASGSFQSASFATRSFTFDDTTAHSEFCFRTRAEK
 RRILVLETSIKLKPDKYATSGHTRRCAIGLLHSII
 35
 >16:04:57|GENSCAN_predicted_CDS_4|1257_bp
 atgaaaccattcccaaggaatccaaggaacccttgccgagtgtagatgctactgttggtcatgacgtgtgtactttgttgccg
 tagagcttgatccctatctcaggagcagcatgggaatgaggacgctagagctcaaagcggcgctctgtattacagctccttgccg
 gttgccgatgaggttttgctgctcatatttgccctgcaagtgtcggcttggtgccacatgtcacatgttgctgcaaccggaatcct
 40 ttcaaggcaactgcaaaagcaaaaggtcaggcggctcagctccactaaaccacagctttgctttcacggctgctgtggctggat
 aattactacaaaggtgaaacgttcaccgaaaactcgccagcatcatgagcgggtttgctggggagcggcatagccttggtgagt
 gcggtggttggtggaacggaacaaatcctgctgattggcaggacattgattggccgcatgagccatactacaaactgattcgacc
 agcccctttgtgtgactgtcactcgaactgtcgggtccaagtgcaaatgtatctgtgtatctgtaggtttctgtgtgcgccgtct
 tgtcagcgcttgacatgaaaatagtttggccaacttgccatgcaaaagcgatttctattaggagccgccatcgccgacatgtgct
 45 gccgaaatcgggtgatttggtgcaactgcagctagatccagtcagccaattgacgaaagagccgacggcagcggtcttgcaact
 ggttaccaaagtatgcgataacaataacatcgccactatgtggtcgttgctgggggttacgggcagtcagtcacgggtcacgggtgc

aaccctccgctccggccaaaacgagtcacagaacaatggccaaggacgaagggggggaggggggattcaataacaaca
 gcaggaacaacaaacattctgctcccacgcaagagcagcaggaactgtggcaaaacagctgctgcaggatcaacgagacgat
 tgcattgccagtgggaagcttcagctcgctcattcgcgagacgcgtagtttcacgttcgacgacacaaccgctcacagcgaattt
 5 tgtttcggactagagctgagaaacggcgaaatttggtgctctggaaacatcgattaactaaaacccgataagtatgcgacaagc
 ggtcacactcggcgatgtgcgataggattgctgcattcgattatag

- Drosophila* Gene Hit** rescue sequence: mitotic heterochromatin fragment clone CH(2)6
 (L36595) and subtelomeric heterochromatin repeats (L03284).
 TBLASTN with ORF1: nebula (nla) (AF147700)
 10 **Human Homologue** BLASTX with nebula: Down Syndrome candidate region 1-like
 protein 2 (AF176117)

***Drosophila* EST** rescue sequence: CK01138 (AA141069)

- 15 **Annotated *Drosophila* genome genomic segment** AE003712
Annotated *Drosophila* genome Complete gene candidate CG6072 - nebula
 CG6046 - sap18

- 20 **Human homologue of Complete gene candidate** CG6072- 8e-36 'ZAKI4 a thyroid
 hormone responsive gene in human
 skin fibroblasts' also known as
 DOWN SYNDROME CANDIDATE
 REGION 1-LIKE 1; DSCR1L1
 EMBL:D83407
 25 protein_id:BAA11911 gi:143504

- 30 CG6046- 3e-45 2108210 (U96915)
 sin3 associated polypeptide p18
 [Homo sapiens] and gi5032067
 C7E479FFE9CA5774
 [refNP_005861.1] sin3-associated
 polypeptide, 18kD [Homo sapiens]
 (1.90E-43)

- 35 **Putative function** Nebula unknown function, Sap18 transcription factor

Confirmation by RNAi Both show reduction in G1 and G2/S peaks indicating fewer
 cycling cells

Line ID 234/50
Category Meiotic defects in testis: cytokinesis defects, abnormal spindles.
(Ab-02/12)
Reversion R
5 **Map Position** 89B

Rescue ID 2C5E
Rescue Sequence
10 GTTTGACTGCTGACGTCGATCGGAACTTGCTGCGCTGACGTACAAAATCGCGA
AGTGAATAAATAATATGGATGAGACTCCTGTTTCGCCGACATATACAATAGTG
CTCAAGACCCTAATGGAATTATACGTTAATAACCAGCCACATTTCTTAGATAT
TTCTAATATGAGCCATCTGCTGCAGGTTCTTTCCAATATCTAATTCTAGATCTT
CTTCGAATACGACCTTTTGGCCATGAAACGATGATTGCCACTTCATTACAA
GCATTAATTTGTCATGATTCTCTTAAGCGTGCACCTTATCTGAAAGTCTGAACA
15 GCTGGCTGCGAAATGGATTCCCCGGATTGGAGATGGCAAGTAAATCTGTCCTC
GCTACAAACAAGTGGGCACCACTGGGCATTCTGGGGAATAGGGATATGGGTTG
GAAA

20 **Drosophila EST** rescue sequence: CK01138 (AA141069)

All other entries as for 1066/5.

Example 3 (Category 1)

Line ID 1104/16
Category Mitotic defects in brain: cytokinesis defect
5 (no overcondensation of diploids, high polyploidy)
Reversion R
Map Position 92A

Rescue ID B5P
10 **Rescue Sequence 1**
CTCCGGACACGCAGTAGCTAAATAACAACTCATTACTAGTATATTACTGCCG
CCGATTTGCAAGCGCGTACCGATCCCGATACCAGGCCAATCGCACTCCCCAGT
TGTACGTCATCACTTAAGTAATAAATCAGCGGCAAATCGCATAAAATTGCTATT
GATATTCCGCCCGCTGTGTGTGCGTGTGTATTGCAAGAGAGTGTGTGTGTGT
15 GTGTGCATATGACTCGTGCCTTAGCCGACAATTGGAGAAAAAGCATTACCAA
TCCCAATTGGCTAACTAACTAAAGTTGGCTTGGCCAAACATAAAACAAAAAGT
GCGGGCGCAGCGATTTGGCAGCGAAACATATACACCAAAGCGCTATTGGCAG
ATATATATGTAGATTAAATATAGAAAGTGCCTGCGAAGGTTAAGAGTCGAGT
GCAAGTGCATTTATATTTGGAAATAATAAATGCTACAAT

20 **Rescue ID** B5E
Rescue Sequence 2
GTCCGGAGCGGAGCTAAAGTTCGATGTTTCGTGCAAAACACTTCGATTCCGATA
GGCGGATGCTATCGATTTTCGGCGATGCCCGTTGGTCACACTTGGTGGTGGGCG
25 CTGCCCCGTCGCCGACTATCGATAGCACAAGCGGGTTATTTAGGTGTGCGCAGC
TTGTAAGGGTGACTCATGCTGTATAAAATTATTATAAAAAGTTAATGAATATAA
TATAGTTATAATAAAATTATATATAAATCTATAAGATCAAAGATCATCAGTTA
TCATTTATCATTTGATTATATGAAAAACAAGAACAGAAACAAGATTTAATAGG
TTTTTGAAATGTGAAAATGTGGGTTACCCCAATTCTTATTCGAAATTAAATAA
30 CCTAAAGAACAGTTATACACAGATAGGTAATTTGCACATAAGCCAAATTTTGT
CTAGAATTCCGCGGAATTAATTCCTGAAGACGAAAGGGCCTCCGTGATACGCC
TATTTTATAGGTTAATGTCATGATAATAATGGTTTCTTA

Genomic hit, Accession No. AC006589
35 **Associated ORF**
Genscan: ORF1 predicted sequences
>/tmp/aaaaainga/GENSCAN_predicted_peptide_2|850_aa
MATRGANVIWFRHGLRLHDPALLAALADKDQGIALLPVFIFDGESAGTKNVGY
NMRMRLDLSLQDIDDQLQAATDGRGRLVFEGERPAYIFRRLHEQVRLHRIEIQDC
40 EPIWNERDESIRSLCRELNIDFVEKVSHTLWDPQLVIETNGGIPPLTYQMFLIRCTH
HNGDVNGDEDTGEGEGTGGRIDWAKEGACWRAGNSDEQECQACQSVSSVIMM
VLQYSNPAHHCQLLECLMTLKHNVVKDILCVVAYGTAVSRTSAKLLFFYYWP
AFNANLFDRKVLLSKLTNDLVPFTCQREHCPNSGNAEAAKVCDHSISIAAYAPDC
PPPLYLCIECANEIHREHGSLEFGDILHPMQQVSMVCENKNCRSNEKSAFSCFSTE
45 CASFNGNHPIRYCSQCHSNRHSRRGGDHVVHRSRSLQPAWQMDPEMQMHMVESV
VSLLREAKPLNFEPGKESSSESCKNGSGITADNISLEERQRLGRYGIWLLVGRCTP

TADTPVEVLGRILSMLFHWFHVTAYSYDGFISCLVPHPEYARVGGHWETLASRT
 SHLKEGLQRLICLVPEYVITSEIWDYVMPHWMEATNDVAEKELNELKIVLSKILD
 PEMSPLGFD AKTMYNFVAIRFEKTTAKVQQQALHWLQILTKLEILPLVQLFAMF
 GDGVRIMKYGIQHELMREKDAQSOSLAKAPKTPCKESKETKADMANPPRPPVVE
 5 DDSGNTSAISDDEAPTNRHTEFSTDAEHNLTCCILMLDILLKQEMELQDVEQHMGI
 HTSVCENVSRLIKCMVTAARVGLSSHVCALKVPIEDIIIEEEKSSRKSPPESDKEKTR
 DRDVSLSMAPLPIPLGPLGGFADP

>/tmp/aaaaainga|GENSCAN_predicted_CDS_2|2553_bp

10 atggccacgcgagggcgcaatgtgattggttcgccatggattgcgcctccatgataat
 cccgctctattggcgcgcctcgcgataaggatcagggtatagccctaattcccgtttcatattc
 gatggagagagtgacaggtacc
 aagaatgtgggttacaatcggatgcgtttcctcctggactcgttcaggacatc
 gatgatcagctacaggcggcaactgatggacg
 tggacgcctcctgtgttcgagggcgaaaccggcttatatcttccgctacatgagcaagtc
 gctgtcacaggattgcatag
 agcaggactcgcgagccaatttgaatgagcgcgatgaaagcatccgttctctatgctgggagct
 gaatacgaacttgcgagaag
 15 gtatcacacacgctttgggatccgcaattgtgattgagaccaatggtggcattccaccgctgac
 ctaccaaatgttctgatacgc
 gcacgcaccacaatggagatggaatgggatgaggtacgggagaaggagaaggaaaccggcgga
 aggatcgaactgactgggcta
 aggaaggggcctgttggagggggaaactccgacgaacaggaaatgcaggcctgccaatcag
 tgcctcggatcatgatg
 gtgtccagctactccaacaatccagcgcacatgccagctcctggagtgctgatgactcttaag
 cacaatgctgtaaggacatc
 ctctgcgttggcatacggaaaccgctgtttcccgccactcggctgccaagctgctcttctact
 actgcccagccttaacgccaatc
 20 tgttcgatcgcaaaagtctactctccaaactaaccaatgacatagtccttcacctgccaacggg
 gagcactgtccgaactccggg
 aatgcggaggcagcaaaagtgtgtctacgaccacagcattagcatcgatacgcgccgattg
 tccaccgccctttacctgtgca
 tcgagtgcccaacgagattcatcgggagcagcgaagcctggagtccgacattctgcatccatg
 cagcaggtatcgtatgg
 tgtgcgaaaacaagaactgtcgtctcaacgagaagtcgccttctccatctgcttctccacg
 gaggatgtgccagctcaatgcaac
 25 catccgatccgctactgcagccagtccacagtaataggcacaattccggcgaggtggcgatcac
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 cgagagcgaagccacta
 aactttgagcccggaaggagtcctcgtctccgagtccaaaaagaacggctccggcatcacag
 ctgacaatatttctgtgagg
 aacgccagagactgggacgctatgtatctggtactgtgggtcgtgtacaccactgcagatac
 cccgtagaagtctgtgg
 caggattctgagcatgctctccactggttcatgtaaccgcttactcatacgaatgtttatctc
 ctgctgtgccatccccggga
 30 gtatccccgttgggagccactgggagaccttggcgtcgcgaacaagccacttgaagagggtct
 cagcggcttatatgctg
 gtgccatagaggttatcacttccgaaatttggactatgtgatccgcactggatggaggccatc
 accaagacgtggccgaga
 aggaactgaacgagctgaagattgtctcagcaaatcctcgatccggaatgtcgctctgggct
 ttgatgcaaaacctgtac
 aactttgtggccattcgattgagaagacaacggcaaaaggtgcagcagcaggcactccactg
 gctgcagatcctaccaagctgg
 agattctcattccactgttcagttgttcgcatgttcggcgatggtgttcgcataatgaatac
 ggcatccagcagcagctgatgcg
 cgagaaggatgcccaatctcagtccttgccaaggtcccaagacccgtgtaagagagcaagg
 agaccaaaagcggaatg
 35 gccaatccgccaggcctcctgttctcgaggatgactctggttaacgtctgccatttcggtatg
 acgagggcgccacgaatcgtca
 cacggaattctccacggatgctgagcacaatctcacctgttgcatcctcatgctggacatac
 tctgaagcaaatggaactacagga
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 agtctcgcgcaaatctccaccg
 40 aatccgacaaggaaaagaccggtgatcgagatgtttccctctcgatggctccactaccattcc
 gctgggaccttaggaggattg
 cagacccttaa

Human Homologue BLASTX with EST: Phosphatidylinositol transfer protein
 (P48739)

45 **Drosophila EST** SD01527 (AI530804), GH18602 (AI387906)

Annotated Drosophila genome genomic segment AE003725

Annotated *Drosophila* genome Complete gene candidate CG5269 – vib PIP transfer protein

5 **Human homologue of Complete gene candidate** 1e-90 1346772 P48739
PPI2_HUMAN
PHOSPHATIDYLINOSITOL
TRANSFER PROTEIN BETA
ISOFORM

10 **Putative function** phospholipid transporter involved in lipid metabolism

Confirmation by RNAi Slight reduction of G1 and increase in G2/M peaks
indicating arrest in G2/M

15

Line ID 418/32
Category Meiotic defects in testis: cytokinesis defects. Dark bands in eyes,
dominant.
Reversion ?
5 **Map Position** 69C

Rescue ID G2E

Rescue Sequence

AGCTAAATAACAAACTCATTACTAGTATATTACTGCCGCCGATTTGCAAGCGC
10 GTACCGATCCCGATACCAGGCCAATCGCACTCCCCAGTTGTACGTCATCACTT
AAGTAATAAATCAGCGGCAAAATCGCATAAATTGCTATTGATATCCGCCCGCT
GTGTGTGCGTGTGTATTTGCAAAAGAGTGTGTGTGTATGTGCATATGACTC
GTGCGTTTAGCCGACAATTGGAGAAAAAGCATTAGAATCCCAATTGGCTAACT
AAACTAAAGTTGGCTTGGCCAAACATAAACA AAAAAGTGCGGGCGCAGCGATT
15 TGGCAGCGAAACAAAAACACCAAAGTGTTATTGGCAGATATATATGTTAATTA
AATATNAAAAAGTGCGTGCGAA

Genomic hit, Accession No. AC006589

20 **Drosophila EST** SD01527 (AI530804), GH18602 (AI387906)

Rest of results same as line 1104/16

Example 4 (Category 1)

Line ID 1285/1
Category Meiotic defects in testis: cytokinesis defects
Reversion ?
Map Position 85D1-5

Rescue ID D8E

Rescue Sequence

10 GTTCGCAAAAAATATATCTCACCGTGAGTGCAGAAAGAGAAAAAGAGAAGCGG
 AGAGGTGGAGAGCAAGTGGACATGAATCGTCGAGAGTCAGAGAGAGAGAGG
 TGGAGAGGGTGAGCAGCTGTTGTCTGACAATAACATAATCAGCAACAATTTAT
 GCTGTTTAAAAAGAGCAAGAGAAACGCTAATGAAGGGGAACACGGGCAGGGT
 CAGGGGTTGGTGGATCCCCACATATCTCTCTTTACCGCCCCCGCTCTGGC
 15 ACCCTCTCTGTCTGCTCTCCATTAGCCGCACACGTGCAAGCTTAGCATTCTATC
 TGTCTGTCTCTGTTTGTGTTTGTGTTGCTAAGCCGAATTCT

Genomic hit, Accession No. CSC:AC014256

20 Associated ORF

Genscan ORF1 predicted sequences

>/tmp/aaaaakfaa|GENSCAN_predicted_peptide_1|702_aa

MIQRCVVLLWIVCFCDLFLGLLFLKRKRNAHTPPPPQFTTYRHLLCYCFRNGEIM
 ANICLSRLSVLEEIVLLLRVPCAFYFVDYYYVPCLLSVLSSEFLYHDQLKVFNRTK
 25 QQHQQQQQQQQQQLYQQHQQQQQQHYGPPPPYFQQLHQHQHQQQQQQQQQQ
 HQQHMKFLGGNDDNRNGRGGVGVGTDAIVGSRGGVSQDAADAAGAAAAAAGV
 YVFQQRSPGGVGVGVGGVGGGVPGVGA VGSTLHEAAAAEYAAHFAQKQQQT
 RWACGDDGHGIDNPDWKWYNPPMNPANAAPGGPPGNGSNGGPGAIGTIGMGS
 LGGGGGGGAGGGNNGSGTNGGLHHQSMAAAAANMAAMQQAALAKHNHMI
 30 SQAAAAVAAQQQHQPQHPQHPQHQHPQHQHPQHQHPQHQHPQHQHPQHQHP
 LGIHPGQQQQQQQQQQQQQQHPGQYNANLLNHAAALGHMSSYAQSGGSMYDH
 HGGAMHPGMNGGMPKQQPLGPPGAGGPQDYVYMGGQTTVPMGAAMMPPQNQ
 YMNSSAVAAANRNAAITTSTAKKLWEKSDGKGVSSSTPGGPLHPLQIPGIDPSS
 VWKDHTWSTQGENILVPPPSRAYAHGGASDTSNSGNAGILSPRDSTCAKVVEYVF
 35 SGSPTNKDSSLSGLEPHLRNLKFDDNDKSRDDKEKANSPFDTNGLKKDDQVTNSN
 GVVNGIDDDKGFK

>/tmp/aaaaakfaa|GENSCAN_predicted_CDS_1|2109_bp

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 45 atggccgaggagcgctggcgttggcacggatgccattgtaggatctcgaggtggcgtctctcaggatgccgccgatgcagctg
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ggtggcgggtgtgccaggggtcggagccgtaggctcgaccttgacgagggccgcccggcggagtagccgcccactttgccc
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 5 atcatcaatcgatggccgtgcagctgcgaatatggcagccatgcaacaggcgccggcggttgccaagcacaatcacatgatat
 cacaggcagcagccgcagttgcagctcagcaacaacatcagcatccacaccagcagcatcccagcagcagcagaacagca
 gcaggcgagaaaccaggggcatccacatcaccttatggcggtggcaatggactgggcaacggcaatggattgggcatacaa
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 10 aatgaacggcgcatgcccaagcaacagccattgggtccacccggagccggagggacccaggactatgtctacatgggtggc
 cagaccactgtgccatgggagccgaatgatccgccacagaatcaatatgaacagctctgtctgttgagctgccaatcggga
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 cgttgcaccccctgcagatccccggcatcggggatccctcctccgtgtggaaggatcacacctggtccacacagggcgagaatat
 attggtgcgccccctcgcgagcctacgccatggagcgctccgatactcaaacagcggaatgcgggcatactgagtc
 15 ccgcgattcgacttgcgcaaaagtgttgaatatgtttcagttgctcgccaccaacaaagatagctcgtttccggattggaacc
 gcatttgcggaatctaagtttgacgacaacgataagtcacgcgacgataaggagaaagcaaacctcctggttgacacaaacggtt
 tgaagaaagacgatcaggtcacaaactcaaatggtgtgtcaacggcattgacgatgacaagggcttcaagtga

Drosophila Gene Hit TBLASTN of ORF1: pumilio protein (L07943)
 20 **Human Homologue** TBLASTX with pumilio: Soares fetal heart NbHH19W Homo
 sapiens cDNA clone (W77820)

Annotated *Drosophila* genome genomic segment AE003681
 Annotated *Drosophila* genome Complete gene candidate CG9755 – pumilio RNA

25 **Human homologue of Complete gene candidate** 1e-154 1944416
 dbj|BAA19665| (D87078)
 similar to D.melanogaster
 pumilio protein (S22026)

30 **Putative function** Putative RNA binding protein which is localised to the cytoplasm.
 Wild-type allele of pum involved in development of the abdomen
 (embryos) and of the imaginal discs (larvae or pupae), perhaps
 35 having a function in signal transport.

Confirmation by RNAi Only wild type profiles observed

Example 5 (Category 1)

	Line ID	1389/1
	Category	Meiotic defects in testis: segregation defect, cytokinesis defect
5		(Ck-09/32)
	Reversion	NR
	Map Position	93B4-8
	Rescue ID	2C9P
10	Rescue Sequence 1	GTTTCGGGGTGTGTGCGTGCTTGCGAGTGTGCCTGTGTGTGTGTAGGAAAGGAG CAAGAAGCAGCAGCAGCGGCAGCAGTAGAAATAGCAAAAGGAGGCAGCAAC AACAAATAAGCTAGAGAAACCGCCAGCAGCAGCCCCCTAATAAAGAGCAGAGA AAAAAATGAGTTCAAGTTGTGAAAGGTGTGTGCCGTTACACTACAACTACAA CACCACCATCAGCGGCAGCAAAGAAATACAACAACAAATACGGCAATCTCCA GACAACGCGAATGTCGAAATTGTGTATACAATTTATTAAGAAAGCAAGAGCA GCAACAACAATGACCAGCTGCAGTTCATCAGCGGTGTCCTCCTGAATGCCGCT GTCGTCGTTGGTGTCTGCCACCGGCGGTTCTCAATAATAAGGGCAGGAGGAG CTGCTTAGGTGCACACAATGTAGTTTGGCTTGGTGAATGCTTCTCTTTTGTG CTGCTGGCGCATACGTTCTCTCTCCCTCATGATCTCAGTTGTCTGCATCGA TGAGCCGCCACCAACGGTGGCTTCTTCTGCTCCTCTTTGGCAACGGACTGCTG CAGTCTTGCCAGAATTTTCTAAATACTGAGCTTCAACTTGGTCTGCTTGGT AATGGTATACCATAAGCCATGGACTTGATGCCCCTACAAAGCTCTGTGATTG AAATGGGATGCA
25	Rescue ID	2C9E
	Rescue Sequence 2	CCCCGAACGCACTTTATATATATAAATATATATATTATTTTCTTTCACCTATTTT CGTTTCGGCCGCGACAGCGAATATGCAATTTTCTCTCAATTGATTTTTTACA CACTCGCACTCCTTTTCACATGCGTGCAGTTTATGTTGCTATTGCTGCTACTGC TGCTGTTGTTGTTATTGTTGTTCTGGCTGCCGCTGCAGTGCAACTTGTAACACT TTCACATTTATGACATAATGCACTGGCCATATTTTGGCTTGGCTCTCCGTTTGT GCAACTGCATGTTCCAGTGCTTTTTTAATATTTATGCTGCAGTGCGTGCAAAT TCGAACGCGAGACGATCCGCTTTTCGCTGCATCTATGCGCTGAAGATGTGCTG CAGTCGATGGGCTCGTCGATAGTGGGAAGGCTCGGTGCCGGCACTATCGATTTC CCAACACCATACGATAATATCGGCTAAAGTTATCAATATCGAAGTTTACTATA TTTCGGGTTTTTACGTTTTAAATCTACCTTATCAACATTTTTGNAAGAAGTAAA AAGTAGTTCTCTTATGGATGCATC
40	<i>Drosophila</i> EST	several including LD10379 (AA816796)
	Annotated <i>Drosophila</i> genome genomic segment	AE003733
	Annotated <i>Drosophila</i> genome Complete gene candidate	CG3421 - novel protein with weak homology to myosin

- 5 **Human homologue of Complete gene candidate** Ensembl predicted
Gene:ENSG00000071333
Clone:AC022505
Contig:AC022505.00011
5.60E-37 (predicted protein
with Core domain in kinesin
and myosin motors
ENSG00000087179)
- 10 **Putative function** Possible novel motor protein involved in cytoskeleton organization
- Confirmation by RNAi** Marked reduction of G1 and G2/M peaks indicating fewer
 cycling cells

Example 6 (Category 1)

Line ID 293/9
Category Mitotic defects in brain: cytokinesis defect
 5 (no overcondensation of diploids, very high polyploidy)
Reversion NR
Map Position 66B

Rescue ID 2G5E
 10 **Rescue Sequence**
 GTACAAACGAATTATTTGTCTCCTTGTGCGTTCGTTTTATTGTGTTTCGAGTTCT
 GTTGGTGTGTGTTTTTGTGTATGTTCCACGAGTTGTTTCGCATTAATAAATAAC
 TGCAGAAAGATCCATGGAAATGGAGACCATTGAAGAGCAATCGAAGTGCAGTG
 AGTACTGAAAAGAGGGCGCGGGGCGTGGCAGCTCCAAATGGCCGGCGAATTTA
 15 TCATTTTCAATGTCGTCCAAAGGGGTTGGGTACGGGGTAAACCACATTCCG
 GGCCAAAAGATCCTCATAAAAAATGTCGCTGCCAGCAAAATGCAAAAAATAAA
 AAAAAATAAGAACGACTATAAGTACATCTTTGTGTGTATTTGTGTGACTAAAA
 AAGCAACGGCATCGTGTGCANATATTTTAATCTTTNTTTCTGAATTTATTTTCG
 GTGTACAAAATATTTATCGCATAAATGCGAAATGCCTCCCTCTCTTCATCATCG
 20 T

Genomic hit, Accession No. AC008303

Associated ORF

25 Genscan ORF1 predicted sequences >20:53:38|GENSCAN_predicted_peptide_3|261_aa
 MMDNDDALLNNGGPQSGAETVYGTEDDNNMVMSEKCRIFPATQRTGFVGFATFSG
 VLLLDLGLALQHCDVIRIDVNIATLEQIKRERQEELAARERJRAQLAADRAEQARF
 NTPDISSTNSVAATAASNVTITDASVSSVDETRLQIRLPGGIQRKTSFPAGEVLAT
 VRVYVRNEMLAASDVRDFTLATSYPREFQTEDEVKTLNELNLVPNAVVLVLT
 30 EQVNPADDQTAKRSASTKRTKTHRHKRQLMADEPQSDHYKN

>20:53:38|GENSCAN_predicted_CDS_3|786_bp

atgatggacaacgatgatgcactgtcaacaatggaggaccacagtcggagctgaaactgtctacggtaccgaggacaacaac
 atggtcatgtcggaagtgcgcataatccggcgactcagcgtactggattgtggcgcgacgttttcgggagtgctgctctt
 35 gatcttggtgccctccagcattgtgatgtgatccggattgatgtaacattgcaacgctggaacagattaagcgtgagcgtcaggag
 gagctggcgccaggaggagcgcaatcgtgccaaattgcagccgatcggcgagcaggcacaacgttttaatacggcgacat
 tagcagcacgaccaattcgggtggcgccaccgctgcctcaacgtgatcacaacagacgcctcggtaggtcggtagcagaga
 cgaggctgcagatccgactaccgggtggcattcagcgaccaaactcttccagccggcgagtgctggtaccgttcgtgtcta
 cgtgcgaacgagatgctggcgagcagatgtacgcgactttaccctggtaccagttaccacgaaggaggttccaaacgg
 40 aggacgaggtcaagaccctgaacgagctaaatctagtgcccaatgcgggtggttctggtgctgaccaaggagcaagtgaatccag
 ctgatgaccagacagcaaacgatcagcaagcacaacgcacaaaaacacagacacaacggcgaattgatggcagacga
 gccacaatctgaccattataaaaactga

45 **Drosophila Gene Hit** rescue sequence: pebble (rho1 GTPase exchange factor)
 (AF136492)
Human Homologue BLASTX with pebble: KIAA0337 (AB002335)

	<i>Drosophila</i> EST	SD09146 (AI542703), SD01796 (AI530981)
	Annotated <i>Drosophila</i> genome genomic segment	AE003557
5	Annotated <i>Drosophila</i> genome Complete gene candidate	CG8114 - pbl pebble rho1 GTPase exchange factor
10	Human homologue of Complete gene candidate	2224615 dbj BAA20795 (AB002335) KIAA0337 [Homo sapiens (3e-21) also mouse homologue 3e-95 42359 transforming protein (ect2) - mouse >gi 293332 (L11316) ect2 [Mus musculus]
15	Putative function	A guanyl-nucleotide exchange factor involved in signal transduction which is localised to the mitotic anaphase. pbl is required for the formation of the contractile ring and the initiation of cytokinesis in <i>Drosophila</i>
20	Confirmation by RNAi	Slightly reduced G1 and G2/M peaks indicating fewer cycling cells

Line ID 542/3
Category Mitotic defects in brain: cytokinesis defect
(very high polyploidy)
Reversion NR
5 **Map Position** 66A
Rescue ID 2A1E
Rescue Sequence
GTCCAGTTAATGAAAGTAAACGAATCGAGTACAAACGAATTATTTGTCTCCTT
GTGCGTTTCGTTTTATTGTGTTTCGAGTTCTGTTGGTGTGTGTTTTTGTGTATGTT
10 CCACGAGTTGTTTCGCATTAAAAAATTAAGTGCAGAAGATCCATGGAAATGGA
GACCATTGAAGAGCAATCGAAGTGCGGTGAGTACTGAAAGAGGGCGCGGGGC
GTGGCAGCTCCAAATGGCCGGCGAATTTATCATTTTTCAATGTCGCCCCAAAGG
GGTTGGGTACGGGGTAAAACCAATTCGGGGCCAAAAGATCCTCATAAAAAA
TGTCGCTGCCAGCAAATGCAAAAAATAAAATGAAATAAGAACGACTATAAGT
15 ACATCTTAGTGTGTATTTGTGTGACTAAAAAAGCAACGGCATCGTGTGCGCANA
TATTTTAATCTTTTTTTCTGAATTTATTTTCGGNGTANAAAATATTTATCGCATA
AATGCGAAATGCCTCCCTCTCTTCATCATCGNTTCCCCTNACTCTCCCTCTCTT
CGCCCGACACTGTACCGACGCAAGAAGAAC

20 **Genomic hit, Accession No.** CSC:AC018042
Drosophila EST SD09146 (AI542703), SD01796 (AI530981)

rest of results same as line 293/9

Example 7 (Category 1)

	Line ID	229/30
5	Category	Mitotic defects in brain: cytokinesis defect. Meiotic defects in testis: cytokinesis defects (Mitotic higher level of condensation, polyploidy, Meiotic:
	Ck05/07)	
	Reversion	?
10	Map Position	91F
	Rescue ID	A7E
	Rescue Sequence	TCTTGGCCAAACAACGCGAGCAGCTGATGTCGCATGGTGGGAAAATGAGGGT GGCGCGAGTGGAAGTTGCCATATCGCTGCGATCACAAGCAGCAAATATGGAA 15 GATTAAGCGGAAAACGAAAGACAAAATAATTACAATCAAACAACCGAATTAT AAAAAGAAAATGGTTTGTCTCCGAGTTCGTTTAAATATGCTTATCTACGTATC AATTAAAAAAACCGTAGAAAGAAATTCACGATTCACCCTAATCTAGCTAAGA CACCAACCAAAAATTTCCGATTTACTTTCAGTTGAAGTTGTTGTTACACACTTT TCTTGTGCGATGTTTTGAAGCGCCCATTTGAAATTGATCATTTGAATGTTTTTCCA 20 AATTACCCACATCCATTACAATAAATTTAAATTGCTTATTATTTGATTTTTACT TGGGAAAATCCCGTTGCCAAATTGGAATTACAATTCCAGCTTGGAATCCGTCA AACTTTACAACATAAACTTATTGTTCTTTTCCGGACAATGCTTCCA
	Annotated <i>Drosophila</i> genome genomic segment	AE003686
25	Annotated <i>Drosophila</i> genome Complete gene candidate	CG6284 - novel protein possible sir2 family of transcriptional regulators/telomeric silencing
30	Human homologue of Complete gene candidate	gi7706710 0268A424791DE5BF [ref]NP_057623.1 sir2-related protein type 6 [Homo sapiens] (1.10E-74)
35	Putative function	Putative transcriptional regulator
40	Confirmation by RNAi	Complete loss of G1 and G2/M peaks indicating fewer cycling cells

Line ID 1104/16
Category Mitotic defects in brain, Cytokinesis defect (no overcondensation of diploids, high polyploidy)
Reversion R
5 **Map Position** 92A

Rescue ID B5E
Rescue Sequence
10 GTCCGGAGCGGAGCTAAAGTTCGATGTTCGTGCAAAACACTTCGATTCCGATA
GGCGGATGCTATCGATTTCGGCGATGCCCGTTGGTCACACTTGGTGGTGGGCG
CTGCCCGTCGCCGACTATCGATAGCACAAAGCGGGTTATTTAGGTGTGCGCAGC
TTGTAAGGGTGACTCATGCTGTTAAAATTATTATAAAAAGTTAATGAATATAA
TATAGTTATAATAAAATTATATATAAATCTATAAGATCAAAGATCATCAGTTA
TCATTTATCATTTGATTATATGAAAAACAAGAACAGAAACAAGATTTAATAGG
15 TTTTGTAAATGTGAAATGTGGGTTACCCCCAATTCTTATTCGAAATTAAATAA
CCTAAAGAACAGTTATACACAGATAGGTAATTTGCACATAAGCCAAATTTTGT
CTAGAATTCGCGGAATTAATTCTTGAAGACGAAAGGGCCTCCGTGATACGCC
TATTTTATAGGTTAATGTCATGATAATAATGGTTTCTTA

20 **Rescue ID** B5P
Rescue Sequence
CTCCGGACACGCAGTAGCTAAATAACAAACTCATTACTAGTATATTACTGCCG
CCGATTTGCAAGCGCGTACCGATCCCGATACCAGGCCAATCGCACTCCCCAGT
TGTACGTCATCACTTAAGTAATAAATCAGCGGCAAAATCGCATAAATTGCTATT
25 GATATTCGCCCCGCTGTGTGTGCGTGTGTATTTGCAAGAGAGTGTGTGTGTGT
GTGTGCATATGACTCGTGCGTTTAGCCGACAATTGGAGAAAAAGCATTACCAA
TCCAATTGGCTAACTAACTAAAGTTGGCTTGGCCAAACATAAACA AAAAGT
GCGGGCGCAGCGATTGTCAGCGAAACATATACACCAAAGCGCTATTGGCAG
ATATATATGTAGATTAAATATAGAAAGTGCCTGCGAAGGTTAAGAGTCGAGT
30 GCAAGTGCATTTATATTTGGAATAATAAATGCTACAAT

other results same as 229/30

Example 8 (Category 1)

Line ID 343/5
Category Mitotic defects in brain: cytokinesis defect
(very high polyploidy, chromosomes entangled?)
Reversion NR
Map Position 75B

Rescue ID C6E

10 Rescue Sequence
GGTTTCGAGTTCGTTTCGGTTTCGGCCTCTCCGTTTCGGCTCTCTCTCGCCATCCC
GCTGCCGCACACATTGGCCTCTCTCTCGCAGCTCCACATTCTGAAGGTGGCTGA
CCGAAATGTGGGTCACGACAATGGCGGGGTCGTTGAACTGAACCACCGCCG
CAGTCGCTGCCGTGCTCGCTGCTCTGCCTCTGCTGACGTCGTTAACGTTTTGGG
15 GCTTTCGGTTACGTAGCTCGTGTGCGAGCGAGAGGGGCTACTAGAGGGACTGC
GACACACAAGTTGTGTGCATTTTTTGGCCCCAAAAATCACAATGGGCACAAA
ATATTATTTAATACATCACATAATTGTTTAATCATCTGGCTGGAAAGTGTTCGAG
TTCATCGAACTGCCAGCGATTGACAAATTGCGATTTTCAATGCGGCAAAAATA
TTTACTCAAGCAAATTGTTTGCACCTTCGTTAATTAGGCGGGGAGTGCCGCCAA
20 ATTGGGTCATATTGCAGAAGTATCCAAGAAGTTGGAGAAACAAGCTGCTTAA
ACATTAATTAACACACACCTAAATGGATACATTTGCTACAAACAATTATAAAT
GTTACCCTTATATTAATTTTCAAATTTCTAAATAATCAA

Genomic hit, Accession No. CSC:AC015427

25 Associated ORF
Genscan ORF1 predicted sequences
MVCAMQEVAAVQHQQQQQQLQLPQQQQQQQQTQQQHATTIVLLTGNGGGNL
HIVATPQQHQPMHQLHHQHQQHQQHQQQAQKSQQLKQQHSAVVKLLESAPIKQQ
30 QQTPKQIVYLQQQQQQPQRKRLKNEAAIVQQQQQTPATLVKTTTTSNSNSNNTQT
TNSISQQQQQHQIVLQHQQPAAAATPKPCADLSAKNDSSESGIDEDSPNSDEDCPN
ANPAGTSLEDSSYEQYQCPWKKIRYARELKQRELEQQQTGGGNAQQQVEAKPA
AIPTSNIKQLHCDSPPFSAQTHKEIANLLRQQSQQQQVVATQQQQQQQQHQQHQQ
QRRDSSDSNCSLMSNSSNSAGNCCTCNAAGDDQQLLEEMDEAHDSGCDDDELCEQH
35 HQRLDSSQLNYLCQKFDEKLDTALSNSANTGRNTPAVTANEDADGFFRRSIQK
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PSKNSTVNQINSKLELGNSEMK

>21:55:09|GENSCAN_predicted_CDS_1|1533_bp
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aattgtttacctgcagcagcagcagcagcaaccgcaacgcaaaagactgaaaaacgaagcagcaatgtacaacagcaacaac
45 aaacacctgcaacactagtaaagacaacaaccaccagcaacagcaacagcaacaacacccagacaacaatagtattagtcag
cagcaacagcagcatcagattgtgttcagcaccagcagccagccggcgagcaacaccaaagccatgtgccgatctgagcg

Line ID 448/23
Category Mitotic defects in brain: cytokinesis defect
(very high polyploidy)
Reversion NR
5 **Map Position** 75B

Rescue ID 2G4E

Rescue Sequence

```
10 GCTGGTGGACGCTGCTTTCATTTCGCAAATTGCTCGTCGTTGGCAGCGGTTGTGC
    AGAGCAAGAAAAGCGCGCGAAAAACCAAGCAAAAAATTAATACAGCTGGAT
    CAAGCGAAAGAGATAGAGAGCAGAGTCAACAGCAACAAATGTTCAATAGCA
    AATGATATCGCATATTTTTGTTGGTGCCAGTGAAGTGAGATCAAAGTGAAGTG
    TGCAATGTTTCCTTATTAGCAAATCGTAGAGCAACCAACAATCGAGAGTTCAAG
    TGTCATTTTGAAGCCAAAAAGCAAAATCTCTAATTCAAATATGGTTTGTGCAA
15 TGCAAGAGGTTGCTGCTGTGCAGCATCAGCAGCAGCAACAGCAACTCCAGTT
    GCCCCAGCAGCAACAGCAGCAGCAGCAGACAACACAGCAGCAACATGCAAC
    AACGATAGTGCTGCTGACGGGCAATGGCGGCGGTAATCTGCACATTGTCGCCA
    CACCGCAACAGCATCAGCCGATGCATCAGCTCCACCATCAGCATCAGCATCAG
    CATCAGCACCAGCAGCAGGCCAAGAGCCAACAGCTGAAGCAACAACAACCTCGG
20 CGCTGGTCAAGTTGCTGGAGTCGGCGCCCATCAAGCAGCAACAGCAGACGCC
    CAAGCAAATTGGTTACCTGCAGCAGCAGCAGCAACCGCAACGCAAAAGA
    CTGAAAAACGAAGCACAATCGTACAACAGCAACAACAACACCTGCAACAC
```

Genomic hit, Accession No. CSC:AC015427
25 **Drosophila EST** GM03519 (A801874)

Other results same as line 343/5

Example 9 (Category 1)

	Line ID	36/1
	Category	Meiotic defects in testis: cytokinesis defects
5		(Ck-04/06)
	Reversion	R
	Map Position	79C
	Rescue ID	A8B
10	Rescue Sequence	GAGTAAAGTAAACTACAGAGAAAAAACGCTTTACGGCGAGAGAACGCTTTAA TTATACTTAATTTGTTGTTAATCAAACGCACAGAGCACACAACACAGAAACAC AAAACACCGCTTGGGAAAAATCTGTAGGTAGANGAAAGGAGCTCACGTTTTT CTGGTGCAGATCGAAATCGGTATCGGGTTTATTGCTTTGCCGGATTGTTACTT 15 CACGTTTGTAAATTGCGTTTCTTTGTTTCTTATTCTCCTGCGCACACTTTGATTT GCGTTTGCAACTCGCAATTGCAATTGGCATTGCTATGCGACAACGCGGTT ATTTCCGGTCCGTTTACTTTTCCAATGGCTTGCCTACACACCGCCAAACTTTGG CTTGAACCTTGGGATATTGGTTGCCCGAATTTCTGANAATTTTTCCTT
20	Genomic hit, Accession No.	CSC:AC013886
	Associated ORF	Genscan partial ORF1: >18:33:59 GENSCAN_predicted_peptide_1 99_aa CICFALLGLLIRRKLMVVFGSTSRKAQSLESRRAKNTSQKIGNQYPKFSQVCGKPS 25 KSNDRNNGSCRIANANCELRVANANQSVRRRIRNKETQLTNVK
	>18:33:59 GENSCAN_predicted_CDS_1 300_bp tgtatctgcttgccttgctggctactcattcgcgaaaattaatggtggttcggttctacgtcgcgcaaggcacagtctctaga gtctcgagagtaagaatacatctcagaaaatggcaaccaatcccaagtcagccaagttgctggcaagccatcgaaaagt 30 aacgaccgaaataacggcagttgtcgcataagcaaatgccaattgcgaattgcgagttgcaaacgcaaatcaaagtgctgcgagg agaataagaacaagaacgcaattaacaacgtgaagtaa	Drosophila Gene Hit rescue sequence and TBLASTN with ORF1: nucleic acid binding protein (mub) (X99340) 35 Human Homologue BLASTX with nucleic acid binding protein: poly(rC)-binding protein 2 (hnRNP-E1) (S42471) Drosophila EST several including LD32520 (AA951799 BLASTN matches nucleic acid binding protein (X99340)
40	Annotated Drosophila genome genomic segment	AE003596
	Annotated Drosophila genome Complete gene candidate	CG7437 - mub mushroom bodies RNA binding protein
45	Human homologue of Complete gene candidate	4826886 ref NP_005007.1 pPCBP2 poly(rC)-binding protein 2

>gi|542853|pir||S42471 (4e-75)

5 **Putative function** A putative RNA-binding protein specifically expressed in the CNS
of *Drosophila melanogaster*

10 **Confirmation by RNAi** Only wild type profiles observed

Line ID 472/22
Category Female sterile
(anaphase bridges, lagging chromosomes)
Reversion ?
5 **Map Position** nd

Rescue ID sau 5'spl

Rescue Sequence

10 GCACGATCNCTAAAGTCTNGCANAGCTAAAAATACATCTNAGAAAATCGGCA
ACCAATATCCCAAGTTCAGCCAAGTTTGCGGTGTGTAGGCAAGCCATCGAAAA
GTAACGACCGAAATAACGGCAGTTGTGCGCATAGCAAATGCCAATTGCGAATT
GCGAGTTGCAAACGCAAATCAAAGTGTGCGCAGGAGAATAAGAAACAAAGA
AACGCAATTAACAAACGTGAAGTAACAATCCGGCAAAGCGAATAAACCCGAT
15 ACCGATTTTCGATCGGTGCGGGCCTCTTCGNTATTACGCCAGNTGGCGAAAGGG
GGATGTGCTGCAAGGCGATTAAAGTTGGGTAACGCCAGGGTTTTCCAGTCACG
ACGTTGTAAAACGACGGCC
ANTGCCAAGCTCTGCTGCTCTAAACGACGCATTTTCGTA CTCCAAAGTACGAAT
TTTTCCCTCAAGCTCTTATTTTCATTAAACAATGAACAGGACCTAACGCCNGT
20 AAC

Rescue ID Sau 5'splac

Rescue sequence

25 GTTGTGATCNTCTTGGTNAATCENNNTTGGAAATCCCCTAANGCTTCCGACAA
TGACCCNGNCNTACNNAGCAAANAATNGNAGNACNNGCNGNTGGNCGTANT
ANCAANAACAGGCCCGCACCGATCGAAATNGGNATCGGNTTTATTTCGCTTTC
CGGATTGTTACTTCACGTTNGTTAATTGCGTTTCTTTGTTTCTTATTCTCCTGCG
CACACTTTGATTGCGTTTGCAACTCGCAATTCGCAATTGGCATTGCTATGCGA
30 CAACTGCCGTTATTTTCGGTCGTTACTTTTCGATGGCTTGCCTACACACCGCAAA
CTTGGCTGAACTTGGGATATTGGTTGCCGATTTTCTGAGATGTATTCTTAGCTC
TGCGAGACTCTAGAGACTGTGC

Other results same as line 36/1

35

Example 10 (Category 1)

	Line ID	459/2
5	Category	Mitotic defects in brain: cytokinesis defect. Meiotic defects in testis: cytokinesis defects: (mitotic: high polyploidy, no diploids, higher mitotic index, meiotic: Ck-01/05)
	Reversion	NR
10	Map Position	66B1-6
	Rescue ID	2D5P
	Rescue Sequence	
	GCTCCGTTTCGAAAGTTGAGAGAGACTTGAAACATATGTTCCGGCGTTGCTAGAG	
	CTGGTCGGCTACCGATAGAAACATCGATAGGTCCGATGTTTTTACTCGTATAT	
15	TGATTCANAGTTTGGCTATCGATGTTTTTAGAGTGCCCGCACATTATCTATTTT	
	CATCTCTATTTTCGTTGGTATTTTTTGTATTTTATGACATTTTCTGACTGCAAAAGC	
	AGGATGGCAACGCCAGATTGCCGCGAAAGTACGTTATTTTTAAATTGGCGCAT	
	TGAATATGAAAAATTGCAGGCACATACAGTTTCTAATAAATAATAGCAATAAT	
	TATTATTTAGCTTGTATCATACGAAGTGCACATTACAGCTACGCATCTGAAAT	
20	AATAATTTTAATATATCGTCTTTTCTCCCATCGATAGAGTTCCGCGCCTATCGA	
	TATATCGTTGATCACCAAATAAATAAACTAAATAACGCCGCAATGGAACAC	
	GCGACGAGTGAATTGAGGGAATTTATCTCAGATCTTGTAATTCCGCACCACGT	
	TGCAATGGTAACATCAATCCGGATCACATCACAATGCTGGAAGGCACCCAGA	
	TCCAGAACAG	
25		
	Annotated <i>Drosophila</i> genome genomic segment	AE003557
	Annotated <i>Drosophila</i> genome Complete gene candidate	CG8038 - novel gene
		ribonuclease P homology
30		CG7892 nmo - protein
		serine/threonine kinase
		involved in eye
		morphogenesis
35	Human homologue of Complete gene candidate	CG8038- 5e-24 4309676
		gb AAD00893 (AF001176)
		ribonuclease P protein subunit
		p29 [Homo sapiens]
40		CG7892- protein kinase
		mitogen-activated 7 (MAP
		kinase)' gi:4506093
		and gi:7706445
		D919050533B3C33A
45		ref NP_057315.1 nemo-like

kinase [Homo sapiens]
(3.30E-174)

- 5 **Putative function** CG8038: tRNA processing enzyme Ribonuclease P protein subunit
 CG7892: a protein serine/threonine kinase involved in cell cycle,
 possibly targeted to cytoskeleton
- 10 **Confirmation by RNAi** Both showed a marked increase in G1 peak indicating arrest in
 G1

Example 11 (Category 1)

	Line ID	623/8
5	Category	Meiotic defects in testis: cytokinesis defects
	Reversion	?
	Map Position	37E1-3
	Rescue ID	2E2E
10	Rescue Sequence	CTACGGGCATTTCGCATGTTCTGAACATCTGGTGTAACAAGTTCTGAGCAGTGT TGCCAACTCTTCAGTTAAACAGTTAAAAATAGCTAAAAAATGTTGACGGTAGC TAAATTATAAAGCTAGAAAAGAAATGATATATGATAAAATAAGTATTTGACT CACAGCATTATTTAAGACGGTCAGATGAAGTTACAAAAATCCTAAATTG 15 GCCCGCTGTATCTAAGAATTAATACCAAGAAGTTGTCATCAAAGGTCGAACTC GACGGAAATTCTACTTTGAGTTTTTAAATTTAATAAATATGTATTTAAATAT GTAAATTTGTTTGTAACAAAAATAGTATATAGTATAGTAATAGTAGTTAAG TAGTTTTAAAAATGGCCAGATCAAAGACTTTTGAGATATGATACTAATCAAAA GTCGAATTCGCGGAATTAATTCTTGAAGACGAAAGGCCTCGTGATCGCCTATT 20 TTTATAGGTAATGTCATGATAATAATGGTTTCTTAGACGCAGGTGGACTTTTCG GGGAAATGTGCGCGGAACCCCTATTTGTTATTTCTAAATACATTCAAATATGT ATCCGCTCATGAGACAATAACCCTGATAAATGCTTCAATAATATTGAAAAAGG AAGAGTATGAGTATTCAACATTTCCGGGCGCCTTATTCCTTTTTTGGGCGGCAT TTGCCTTCCTGTTTTTGTACCCAGAACGCTGGTGAAAGAAAAGATCTGAAGA 25 CAGT
	Annotated <i>Drosophila</i> genome genomic segment	AE003662
	Annotated <i>Drosophila</i> genome Complete gene candidate	CG17559 dnt - doughnut protein tyrosine kinase
30	Human homologue of Complete gene candidate	Homo sapiens RYKreceptor tyrosine kinase GDB:21773
	Putative function	growth factor transmembrane receptor protein tyrosine kinase involved in cell growth and maintenance
35	Confirmation by RNAi	Only wild type profiles observed

Example 12 (Category 1)

	Line ID	629/14
	Category	Meiotic defects in testis: cytokinesis defects (Ck-06/09)
5	Reversion	NR
	Map Position	64D
	Rescue ID	2A9X
10	Rescue Sequence 1	GACGGGAGGAAGTAAGTGGGAGGAGAGAGTAGTGCCTCTTTTTTACTGGAGA AATGGACAAACTCTGGGAACTGCGAACTGCGAACTAACCGAGGCAAAAATTG AGAAGCGAGCTGAAAGCGGAATTCAAACAACGCAGCGCTGACGGCGACGCCG GCAGAAGCAGCGCCGCACAAGGCATGCGCACAGAGAGTAAGAAAGAGCGCG 15 GCTAATGAATGAATGAACGAGGCGGAATGCGGGAAGAGCGCAGAGAGGCGC AATGACAAAATAGTTGTAGAAAAGCGCCGGCAAGCGGAACTCCACACTCTTT CTCACTCTCTCTTTCCACCCACACCCCTAGTTCACCGGAAAAAGAAAATTCGTT TGCGGCGGGGGTGTATTTTTCACCAAAAAGAGAGTGTGTGCAAAACGCTAGA GAGAGAGAGAGAGAGAGAGAAAGAACTGACGTCAGTTCTGCCTCCGTCGACGCC 20 GCTGCCGGCGTCCCAAAGCGCCACCACCAAAAAAACGCGAGAAGAAGCAGA ACAAACACACACAAAAATTGCGCACAGTGGAGCAGAAATCAAGC
	Rescue ID	2A9E
25	Rescue Sequence 2	CTCCCGTCGTTTTGAGATCAGCTGCTCTCGCAACAACAACACTATAACTGTA GTTACCGTCTCTTTTGCATCGTTCGTTTTTCGTTTGTGTGCGCAAGTGATTGTGT GTGTGCGTAAGCTTAAAGCTGACTAACAAAACGAAACAAGAAAAATATAAA TTATAGGAAAATTGTTAAATTATAACCAGAAAAGAGAGCGGCACTTACGTGTGT TATTGTGTGCGTGTGCTTTAAAAAGATATAAAAAATAGCAATAGAAAGTTATTA 30 AAGCGTTGGCAAAAAAGTCCAACGAACAGCGAGAGGAAGCGGAGAACGAAA TAGTTAAAGCCAAAGTCGCTGCCGACGTGCGCACTTGAAAACGTGCGCAAAAGTT TGTAACACACCAGTGTGTGTTCGTGTGTGTTTTGCCGGCGTGCCAGTGTGCG TGCGCCTAGAAAAGAGTAAAGAAGCAGAAAGAAAAGGAAGAAGCCGAAGAAG CAGCAAAAGAAGCCGACAGCAAAAAGTAAATAAAATCAAATGCCCCCTGGCA 35 GAATAATATTAAATTAAGACACATACTCAAATTAATAAC
	Genomic hit, Accession No. CSC:AC015076	
	Drosophila EST	LP08767 (AI295205)
40	Annotated Drosophila genome genomic segment	AE003567
	Annotated Drosophila genome Complete gene candidate	CG10668 - novel with homology to ssDNA/RNA binding proteins
45	Human homologue of Complete gene candidate	CG10668 - 3e-12 4506449

ref|NP_002889.1|pRBMS2|
RNA binding motif, single
stranded interacting protein 2
>gi|1082

5

Putative function Possible single stranded DNA/RNA binding protein

Confirmation by RNAi Slightly increased G1 and reduced G2/M indicating G1
10 arrest

[illegible]

ggaagaaagcggaagggtgtggaatggagtgagtgaggagtgaggatggtggcgatgtagtgtgtgctctcgagtgtagc
ccacagttgacgatgatgatcatcatggtggcgccacttgcagcgctgtgtgggaacaccttcagcgctcatccgacttaactgc
ttaatcaaccgaagaagatgaggtggaacttgaagttgaggttgcatggcaaatgctcgagctgctgatctcggtgatctca
atgcaccttaattgtgccttatgaaatgaaaacgatgaagacgatggagagcgtgatcgggtggctccctgtaccaaccgactgc
5 tctctcggttctttgtttgcttggtgtattcttcagctgctgatgtgtgtgtgctgctggcaactgtaaaagcttagcccatggcgctg
acgtcgactgcgacagcgacgctagccgaggcagtgactgcgacgttgccacttttcgcttcgttgcgtgtcgtttcaagtgtc
tctcgtgtcctaaagcgcgcgacgcgaacgctctgaaatcccaagtacaaacgacaacatcaagcagcagacaacaacagtga
ttcgctggcaacaacaacaacaacaacatatattttgtgtatcaattgtcggcctaa

- | | | |
|----|---|---|
| 10 | <i>Drosophila</i> Gene Hit | rescue sequence, ORF1 and genomic sequence: Canton S E78B nuclear receptor superfamily protein (U01088) |
| | <i>Drosophila</i> EST | LP11082 (AI296953 similar by BLASTN to U01088) |
| | Annotated <i>Drosophila</i> genome genomic segment | AE003593 |
| 15 | Annotated <i>Drosophila</i> genome Complete gene candidate | CG18023 - Eip78C
Ecdysone-induced protein 78C
nuclear receptor NR1E1 |
| 20 | Human homologue of Complete gene candidate | CG18023- 4e-32 119100
P20393 EAR1_HUMAN V-
ERBA RELATED PROTEIN
EAR-1
>gi 1082832 pir A32608 |
| 25 | Putative function | ligand-dependent nuclear receptor , putative transcription factor |
| | Confirmation by RNAi | Not done due to failure of PCR procedure |

Example 14 (Category 1)

	Line ID	876/2
	Category	Meiotic defects in testis: cytokinesis defects
5	Reversion	?
	Map Position	73A
	Rescue ID	2H1E
	Rescue Sequence	
10	GATCAACAGAAAATCCAAAAACGAACAGCGCGCGGCGAACGAGAGCCGTT	
	GAAGCCGGCAGAGAAGTGCCTGCTCGCGTCGCTGCCGGTATGTGCGTGTCTG	
	TGCACTGAGAGAAAATGCTCGATTAAACAGAGAAAATTAATAGTAATATAAAA	
	AAAAAAAAAAATTTGTTTATTATTCTCAATTCAATAAAATGTAATTATTTATTAT	
	ATTGGTTGTATAAGAATTTTATAAAGTAGTATAAAATTTTCAATCAAATAAAT	
15	ATGTACATCTAACAAAAAATGTTATTATCTTATAACAAAGAGGTAAAATCATA	
	AGTAGTACGAAATCTTTAAAAGAGAAAGTGTGTTACGCAAAAAGTATTCAAA	
	GCAGTCTTTTATTTAATTTAATTAATTTATTTGTGCTTTATCCCTTATATATATA	
	TGTACATTTCAATTAAGCTAATGGTATAATTAGGTATTTACAGTGTTTAGCTAA	
	GGCTTTCATCTGAAATATTTATTAATTATGTCTAGTTGACCTGTTTTTAGTTTTT	
20	TTGNATAACAATTTTATTATTTATTAAGGAAAAACAAGGGGAGAAGAAAAAC	
	CTTAATTGAAGCAAAGCAGTCTTTTGAACCCACTGGTG	
	Genomic hit, Accession No. AC005633	
	Drosophila Gene Hit rescue sequence: argos (M91381)	
25		
	Annotated <i>Drosophila</i> genome genomic segment	AE003527
	Annotated <i>Drosophila</i> genome Complete gene candidate	CG10162 – Egf2 translation facto
30	Human homologue of Complete gene candidate	CG10162 - 4e-11 181969 (M19997) elongation factor 2 [Homo sapiens]
	Putative function	Translation elongation factor
35		
	Confirmation by RNAi	Not done due to failure of PCR procedure

CATEGORY 2: FAILURE TO ENTER M-PHASE**Example 15 (Category 2)**

- Line ID** 1216/12
Category Meiotic defects in testis: no division
 (no meiosis)
Reversion NR
Map Position 82F1-2
- Rescue ID** 215X-1
Rescue Sequence 1
 AAACCAAGCAACAGAAATATCTCCAGTAGAGAGCGCCACTGGAAGATCGGAA
 TTTTAGTGCTCTGCTCTGACTAACAGGTTTAGTAGTAGTGCTTACTTTTCTAC
 TACGATTTTTGTGCGGGCTAACAAATCTGTTTTCCCACTCCCTCTCTCAGTTTTT
 GCATGGTAACTTTTCGGTCATTGTACTGTTGTTGTTGTTGCTTGACACCCGCAAGA
 GAACAACAACAATCGGAGAAACACTGATAGCGCGGTACAGTGGGGCAGGCCA
 AACTAGAACCTATACATTTAAGATGTCTCCAATTTGTGATTTTGCCTTTCAAGC
 ATACTAGTTCATAGTTGATTGTTTTGTTATGTTTTGTCTTGAATGCGATGTTTCA
 AGAAATCTTATTTTCGAATTACGATATTATTCTTATTCTTTGACTTATTAATA
 TAAATGAAAACGGCGAGTAGAGCAAAAGAGCGACCACTGTGGCTCCACAAGC
 TCGTTTCTCTGTTTCTCATTGCGGCCAGCTCCAATTTGCGCTTATTCACACACA
 CACCTCACTGCTTGCGACTGCAAATTTGTGCAGCTGAACTTTG
- Rescue ID** 215E-1
Rescue Sequence 2
 CTTGGTTTATCACCCCTCTCTCTCTCTATCGCGCGCGCGCGCTCTTTGTGGAA
 ACAGGTATAACTGTTTGGCGTGAGGGAGCACGAAACTCCAGTGGAGACTTCTC
 CGCATCGCCAGCGAAACAAACGATCAAAATGAATACTCTGATAACGTGTGAA
 GGTGAGCAACAAAATAAAGTATAAGAAAATACCGCCACGAAAACAACAACA
 ATAGAAATGTCGACGCACCCTTTTCTTTTCTCGCAAAGAACGAGGAAATGGA
 GAAGCGCAAAACCACATCCCGCTTAAAGAGTCCCTTTCCCCCGCTGGAAGTGG
 AAGGAAAGGCAGCTTAAAGAGGAGCGGGTGGCTTCCAGTATGTGGAAAACAA
 AGCAGACGCCATTGGAATGCCGTCGTTTTTGTGTTGCTAAGCCGGACATGG
 CAATTGTTGCTTTTGTTCGAGAGGGGGTGGTGAACTCATAAATATCAGCT
 ATGGCGAGGGGGTGGGGGCAGTCTTTGTCTGACGTACCGTACTTTTAATTTCTT
 GTCGCCCGGTTTAATCCAATTTATCCAGCTTTGAATTTGCGCGG
- Genomic hit, Accession No.** AC007532
- Annotated *Drosophila* genome genomic segment** AE003603
Annotated *Drosophila* genome Complete gene candidate CG1116 - novel
Human homologue of Complete gene candidate 2495728 HYPOTHETICAL
 PROTEIN KIAA0258(aa)

Putative function No homologies which indicate function

Confirmation by RNAi Slight loss of G1 peak

Example 16 (Category 2)

Line ID	1344/15
Category	Mitotic defects in brain: no mitosis
5 Reversion	NR
Map Position	83C
Rescue ID	2F6E
Rescue Sequence	10 AGCGGGAGTGAGCCGAAAGAGAGTAATTTTGGCCGTCACCAAAAAAGTGGCT GCATAGTGCCAAACCAATGTATGGCCGTTACGCATCTTGTTATTCTAGTGTCTT TGGCTGTAATCAGTTTGCAGTGACAGAGGAGTTCAGTTTCAGTTGACTCGGCT TGGTTCAGGGTTTCTGATTGCCGTCCTCTTCTCCCTCTTCGCCTACAAGTCCGC TGTTCCGGCACCCTGACGTCACCTAGACTTACACCCCTAATCAAAGATCCACTA 15 GTTTAGATTTCTGTCATCAACGCCATATTAACCTTTATAAGCAGTCGTTATATCT CAAGTAGGCAAAAAAGTGTAATAGATATGTATCTAAATTGTCGTACATTCTAT TTATTAAAATTCGTTTTTACATCCAACAGGTGTTATTTTTGAAGTCTTAGATAA CAAACAATATTCGAATTATGTGGTAGAATACTTAGCAATATACGCACATACAT ATACATATGAACATTATATCCAATGCTTTAAAACCGGAATATCAAGACAACAT 20 AATGCAACATCTGGTCCGAGCTATCCAGGCAATCACATTTTGAAGTTCCTCC GGTTATCACACATATATCGATCATACCCCGAAATGTGTAACACAGATACAGCT CACCATCCCTCTGATAAGATCTTATCAAGTTCGGGCTTGCTCGCTATCGTGAAT TGGGTTGAAGGGTCCGCGATAATTGCATTGGGCATGCCATTGGTAATCACAAT TGGCTGATAATGCTGCTGCTGCAATTCCACGGGTATGAA 25 TTCATCAATTGGTTA
Annotated <i>Drosophila</i> genome genomic segment	AE003602
Annotated <i>Drosophila</i> genome Complete gene candidate	CG1347 - novel protein with myosin homology
30 Human homologue of Complete gene candidate	1503990 dbj BAA13194 (D86958) KIAA0203 similar to mouse CC1.(aa)
35	Putative function similar to coiled coil protein with ubiquitin like domain
Confirmation by RNAi	Marked reduction of G1 and G2/M indicating fewer cycling cells
40	

Example 17 (Category 2)

	Line ID	703/16
	Category	Meiotic defects in testis: segregation defects, meiotic failure (Mf-07/75)
5	Reversion	R
	Map Position	83B
	Rescue ID	2E7E
	Rescue Sequence	
10	AAGCAGCCCAACAGCTACGCAAAAAGTTACTTATATTTCGCAGCAAAACAGAT	
	TTTTTGTTTTAATCGTAAGTATAGGAGTGAAAAATAGCGCTAGAGTAGACCT	
	AAGTACACAGAAAGACAAATAGGGCGAGTAAATCGCGGTCCTGGTCATTTT	
	TCTGGCCTTGACCAATCCTTTGTCTGCGCTTTCGTTGGAAAAGGGGTATGTAC	
	GAACTGCGTGCGTACCTAAGGCCAGATTAGTCATCGGGCAGTCATATATTCAT	
15	GCAAAAAATCATTTGGTGGCCGTCGGCCTTTGTTGCGACTGTACCTTGCTCATT	
	TTTAATAAGCGCGACAGCAATATACACACTTTGAACCCCATCCCACATTTTTT	
	CTCACCGTTTCCCCCTAATTTTCGTTTTCCCTGTGCCCATCATTCCGCTTTCGCC	
	ATGTCAGTGTATCGCTTCAAAATGGCGCCGAACCACATGTCTTCGTTCTCGGC	
	TCGTCCGCTTCGTTTCGTGCGCTCGTGTGTCGTCTCATTTCGCTCTCCGAATTTTCG	
20	TTTAACAAAGTGGTGCAGAGAGGGGCCGCTGGATTTCGAGGCAACAACAC	
	ATATACCTA	
	Genomic hit, Accession No. CSC:AC013960	
25	<i>Drosophila</i> EST	several including LD15903 (AA440858), GH20091 (AI389018).
	Annotated <i>Drosophila</i> genome genomic segment	AE003602
	Annotated <i>Drosophila</i> genome Complete gene candidate CG2922 – novel	
30	Human homologue of Complete gene candidate	286001 dbj BAA02795 (D13630)
		KIAA0005 [Homo sapiens] also
		NP_054757.1 HSPC028 protein
		[Homo sapiens] e-179
35	Putative function	Weakly similar to a region of human and murine EIF4G2 translation initiation factors; may act as a translation initiation factor
	Confirmation by RNAi	Only wild type profiles observed

Example 18 (Category 2)

	Line ID	741/3
	Category	Meiotic defects in testis: segregation defects, meiotic failure (Mf-05/31)
5	Reversion	NR
	Map Position	88D
	Rescue ID	H6E
10	Rescue Sequence	GCCTGGAGCCACCTCTAGAGCCACGGCCAAAAAATTGTGTGCCAAAAAATCG TATGGCGTTACGCATCTTGTTATTCTAGTGTCTTTGGTTCTACAAATCTGGCCA ATGGGATGGACGGATTTTGGGGCTTTTGCGCCCCACATATGTNTCTTACAACC CACTCGGCCCGGCAAGTGGGTGTCAATTACGGACATCGGCAATCCGAAGACC 15 GGAGACCCAGAGACCCTCAGACCCCAGGGCCCCATTCGATTCGATTTGAGTT GCGTGGGCGGATCTCACATTAGTCACATCGAAGGAATGAAATAAAAAGAAAA AACATGACGGCCGAAAAGAACTTATCCATCTTCAAAGCTCTCAGAAAATACA AAAAATAAAAACTTTTGACTCTTCGTCTTTCACATTCGAAATCACAAAAT GTCTGCCATAAATTCCAAAGTGAACAATTGAAATAAATTTTGCGCCATGAAC 20 ACGCCGACTG
	Annotated <i>Drosophila</i> genome genomic segment	AE003705
	Annotated <i>Drosophila</i> genome Complete gene candidate	CG12600 - novel protein
25	Human homologue of Complete gene candidate	CG12600- 5e-27 4240227 dbj BAA74892.1 (AB020676) KIAA0869 protein [Homo sapiens]
30	Putative function	putative cytoskeletal structural protein
	Confirmation by RNAi cells	Reduction of G1 and G2/M peaks indicating fewer cycling cells

Example 19 (Category 2)

5	Line ID Category Reversion Map Position	773/1 Meiotic defects in testis: cytokinesis defects, meiotic failure (Mf-02/15) R? 83F
10	Rescue ID Rescue Sequence	2D9P CCACCGCCCATGCCGCCATTTATTGAAAGGCCTGTACGCAGTTTGTTTTGTTT TTCTCTTTTTTGCTAGCTCAAACACAAAATTACTTTTTGTGGCTTGACTGGTGA GGTCTCTCTATCTCGCTTTTTCGTCTTTACCTCGCTCTCATTCCCTCTCTATCTG CCCTGCTTCCTCTCACTATCTATCTACAACGAGGTCAACAAAATAAGTGCGT 15 AGTCAAAAATGTAATTGAATTGATTGACAAACACAGCGAACGTAAATTTCCGT AATGTTTAACCTTGAATTCAAATGAACAACTGTATAAATATAATACACGGGT AAACTCCATTTCAAAGCAAGCTAAAACATTTTAAATACATTTTAGGGAAACGG CCAATTAAAAGAATAATATTGTGGGGATCAATCTGGGGAAAAATGCAGTATC AGTAATGCTGAATATTTATTTTACTAAATTACAATGAAATGTCTCAAACAAAT 20 GGGTTAATCATTTTCTTGCTCCATCTGCTTTTCCCAACTGTATCCAAGTACAAC TACAGCATTATCCTCAACTG
25	Annotated <i>Drosophila</i> genome genomic segment Annotated <i>Drosophila</i> genome Complete gene candidate	AE003675 CG10272 - novel protein
30	Human homologue of Complete gene candidate	CG10272 - 2995577 AC004490 (AC004490) R29381_1(aa) protein includes HMG-I and HMG-Y DNA- binding domain (A+T-hook) found in HMG non-histone components in chromatin
35	Putative function	Chromosomal protein
	Confirmation by RNAi	Loss of G1 peak indicating arrest in G2/M

CATEGORY 3: METAPHASE ARREST**Example 20 (Category 3)**

5
Line ID 1067/13
Category Mitotic defects in brain: prometaphase arrest
 (overcondensation, polyploidy, scattered chromosomes with
 bipolar spindle)
 10 **Reversion** NR
Map Position 69C4-10

Rescue ID 2F8E
Rescue Sequence

15 GTTTGGGCACAGGGTTGTATTTCAATTTATTTTTGGGGGGAGTCGATACGCTCTC
 TTGGCGTGGTCGAACGGTCACACTGGCCGAGAGATAACGGAAAATGTTTCAA
 AGGTAAGTAAAGATTATAAACGTATTAAGCTTAATACTATAATTAGCTTACTA
 TTCCAAGTATGTATAATTATTACACGTTTAAAAGGCATAACGTTAAGTGTAAC
 CAAATTATATCAATGGATTTTGAATACCAATATTATTTATTTTATATTTTGAGC
 20 TTAATATATTTAAATCACATATATTTAAGCCTCTTTATATATGTAAATATTTTAA
 TTTTATTTAAATAAATTATATATTGTTTTGTAATATGATCGAGGGCTGCCACCT
 TGTGATAAATGCTTACCAACACTTTTAGGTACGCCGTTTAGTGACGTAAGTTG
 CGTACCTAGATATCCAGCGAAATCAAAACATTGAGTAAATCGTGGAATG
 ATGAAAATAGCTTAATCTACGGACTCGAACTGCAGGCGCGGGCTTTAACACCT
 25 CAGTACGGAGAGAGCAACGATGTGTGCTTCTTCATAGCCACCAACTCCTTGAA
 GCCCACCAATCAGGTTCACTTAATCCAGTACGAAGA

Genomic hit, Accession No. CSC:AC020333

30 **Associated ORF**
 Genscan: ORF1 predicted sequences: >16:51:11|GENSCAN_predicted_peptide_2|178_aa
 MAQNISPEQSGGAGGGGSKHSDDSMPVKDNHAVSKRLHKELMNLMMANERGIS
 AFPDGENIFKWVGTLAIPRNTVYSGQTYRLSLDFPNSYPYAAPVVKFLTSCFHPNV
 DLQGAICLDILKDKWSALYDVRTILLSIQSLLEGPNNESPLNAQAAMMWNDQKEY
 35 KKYLD AFYEKHKDT

>16:51:11|GENSCAN_predicted_CDS_2|537_bp
 atggcgcagaatatcagccccgagcaagtggtggagcaggcggcggcgcagcaagcacagcgatgactccatgcccggtg
 aaagacaatcacgccgtgagcaaaagactgcacaaggaactgatgaacctgatggccaacgagagggcactctcagcgtt
 40 tccggacggcgagaacatctcaagtggtgggcaccatagcgggtccacggaacacgggtgattcggggcaaacgtatcggtt
 gtcactggattttccaattcctatccgtatgcagcaccggtggaagttcctgacgtcctgcttccatccaatgttgatctgcagg
 gcgccatctgtttggacatactgaaggacaaatggtcgccctgtacgatgtgcgcaccattctgcttccatacaatccctgctgg
 gcgaaccgaacaacgagagtcactgaatgcgcaggccgcgatgatgtggaatgac

- Drosophila Gene Hit** TBLASTX with ORF1: poor homology to several sequences including homolog of RAD6 (DHR6) (M63792), bendless (L20126) and Ubc D1 mRNA for ubiquitin-conjugating enzyme (X62575).
- 5 **Human Homologue** TBLASTX with ORF1: ubiquitin carrier protein E2-C (UBCH10) (NM_007019.1) and ubiquitin-conjugating enzyme E2B (RAD6 homolog) (NM_003337.1).
- Annotated *Drosophila* genome genomic segment** AE003541
- 10 **Annotated *Drosophila* genome Complete gene candidate** CG10682 – vihar ubiquitin-conjugating enzyme
- Human homologue of Complete gene candidate** gi5902146
0B6F58A1F0665D9A
15 |ref|NP_008950.1| ubiquitin carrier protein E2-C [Homo sapiens] (2.50E-50)
- 20 **Putative function** Cyclin specific ubiquitin conjugating enzyme
- Confirmation by RNAi** Complete loss of G1 and G2/M peaks indicating fewer cycling cells. Immunostaining shows metaphase arrest with condensed chromosomes
- 25

Line ID 1105/1
Category Male sterile, Female sterile, Mitotic defects in brain: prometaphase arrest
 (Overcondensation, polyploidy, fewer anaphases, high mitotic index, scattered chromosomes with bipolar spindle)
Reversion R
Map Position 69C

Rescue ID A5B
Rescue Sequence
 GTACATATAATCACAATTGAGAATCGAAAACCCGACCGCCACGAAGCGCGCT
 AAATTACACGCACATACTGAAAGCCAAACAGCGGATAGCACTAGCATCCTAC
 ATATATAGACGTAGATATATAGTCATGGCGCAGAATATCAGCCCCGAGCAAA
 GTGGTGGAGCAGGCGGCGGCGGCAGCAAGCACAGCGATGACTCCATGCCCGT
 GAAAGACAATCACGCCGTGGAGCAAAAGGTGAGTATCACATGGTGCAGCCTA
 AGATAATCCGCCAATATACACACACTCACACTCACCCACAGACTGCACAA
 GGGAACTGATGAACCTGAATGAATGGGCCACCGAAAAAAGGGG

Rescue ID A5E
Rescue Sequence 2
 ATATGTACTGTATAGTGGAATTTAGTTTGATCGGTCTGGAATACGCGTCTGTT
 GCTTTTTCAGATATTTTTTTTTCACTTTTGTGTGAAAACAAAATGGAAGGAGA
 ACGAGAAGAACTGTGTTTGGGCACAGGGTTGTATTTCATTTTTTTTGGGGG
 GAGTCGATACGCTCTCTTGGCGTGGTCAACGGTCCACTGGCCGAGAGATAA
 CGGAAAATGTTTCAAAGGTAAGTAAAGATTATAAACGTATTAAGCTTAATACT
 ATAATTAGCTTACTATTCCAAGTATGTTATAATTATTACACGTTTTAAAGGCA
 TAACCGTTAAGTTGTTAACCCAAATTATATCAATGGATTTTGAATACCAATATT
 ATTTATTTTATATTTTGAGCTTAATATATTAATCCACATATATTTAACCCCT
 TTATATATGTTAAATATTTTAATTTTATTAAATAAATTATATATTGTTTGGTTA
 AAA

Genomic hit, Accession No. AC007328 69B-69C

Associated ORF
Genscan: ORF1 predicted sequences
 >/tmp/aaaaanjda|GENSCAN_predicted_peptide_1|357_aa
 MGKKAKHKKKKGKPEKTAMKADKKQAAARQKKMLEKLGEANIADIQLLEAKEG
 KIEAISESVCPPTPRSNFTLVCHPEKEELIMFGGELYTGKTTVYNDLFFYNTKTV
 EWRQLKSPSGPTPRSGHQMVAVASNGGELWFPNFACISRNQSWFVFHNCRLKAA
 SREKVLLNFNGTVLHPANNIIVHVKLFKKANGFKPWLLDVKLDACRFVRTNFHFP
 VRIIFDLFKDFSTINHTCPYVVLRSRMYTVRRSPRLVHPIVDVPAGHTRPRRKA
 AV
 RGIGCAHRCPLIRMATPCRTNVMMTMLMRGSVRSRVMAICCYRRPAIAIARRRHP
 TAIHSQEVAERLGGLLYPDIQRTNP

>/tmp/aaaaanjda|GENSCAN_predicted_CDS_1|1074_bp
 atgggcaaaaaggccaacacaagaagaagggaaggccgagaaaaaggccatgaaagggacaaaaagcaggcgg
 cgcggaagaaatgctggaaaaactgggagaagcaaatatagctgatatcatccaattgctggaggccaaggaggcgaag
 attgaagccatcagtgaatccgtttgccgcccaactccacgatccaatttcaccttagttgccatccggaaggaggagctc

atcatgtttggcggcgaactgtactggcacaaaaaccacagtgtataacgattgttctttacaacaccaaaccgtcagtgagg
aggcagctgaaatcgccatcgggacccacgcccagaagtggacaccaaagggtggctgtggccagcaatggaggagaactct
ggttccgaactcgcctgtataagtcgcaatcaatcctggttgtgtccacaattgtcgtctgaaggcggccagtcgtgagaagg
cttactcaacttaatggaacggttctacatccggccaataacataatgttcacgtcaagctgttataaaaggccaacggtttaagc
5 cttggtattagacgtaaaactcgtgcttgcgtttgtcggaccaacttccatccgtttgtacgcattatattcgatcttcaaagat
ttctccaccataaaccacacgtgcccatatgtggtcctccgatcgtcgggtatattgtccgccgatccccacgacttgtcacccc
atcgtagatgttccggctattgggcacactcgcctcgacggaaggccggttcgtggcatagggtgtgctcatcgtcgcctct
gattcggatggcgactccgtgtcgtaccaacgtggtgatgatgacgctgatgaggggctcgggtgagatcgagggtgatggcgatt
tgctgctaccgccgacccgccattgccatagcccgctcgcgccacccactgccattgccactcccaagaagttgctgaacgc
10 ctcggtggtcttctttaccggacattcagagaaccaatccgtag

Drosophila EST several ESTs including LD04777 (AA201675)

All other entries as for 1067/13.

Example 21 (Category 3)

	Line ID	1407/13
	Category	Mitotic defects in brain:
5		(weak overcondensation, metaphase with bipolar spindle)
	Reversion	NR
	Map Position	92B1-3
	Rescue ID	2D3P
10	Rescue Sequence 1	ATCACGAATTTGACATTGCTACCAACATTCGGTGCGTGGACTCTGAAAGCTCTG AGTGTTTTGTATGCAAAGCTTTTTTGGACTATCGCGTGGTAAGTAGCCGAAA GAGAAAGCTCTCTTATACGGAAGATGAAGAGTGTGATTCATGAAAATGTATA AGAACGCGGGTCCAAAAAGTCAAGGGAGTTCTAGTGAAATGAAAAGTTCCAA 15 AGGTTTTGAAATCGTTTTATTTTCTCGTTTCGTATAATTATTGGGTGTCGATCTTT GTTGGGCAGTGTAAGCACAAACTTTGAGCTTCATCATACATATCATATGTAA AGCCGGGACGAAAGCTTATGATTCTGTTAAGTGTCCGCCCAAGATAACATTTT TCCAGCCCTTCAAATCTTCAAATAAATACGGCTTAAGGCGAGCAAATTTGTAA ATCAAATGATTTGTAAATAAACATTATATGTATTTTATCATGCCAGGTTAGAA 20 CACATTGTGCTGATGCAAATAAAATTCCAATTAACGCCCTGAATGGGAAGA TGACGCATCTTTAATGGGAATATTATGGTAAATTTAATA
	Rescue ID	2D3E
	Rescue Sequence 2	25 TNCGTGATTATCAGCGTTAATTGTACAATATTATGATTTATTCGAGCTGTAAAT CTTACAGCAAGCACAACTGTAATTATACCACTTAGAATTCGCGGAATTAA TTCTTGAAGACGAAAGGGCCTCGTGATACGCCTATTTTTATAGGTTAATGTCAT GATAATAATGGTTTCTTAGACGTCAGGTGGCACTTTTCGGGGAAATGTGCGCG GAACCCCTATTTGTTTATTTTCTAAATACATTCAAATATGTATCCGCTCATGA 30 GACAATAACCCTGATAAATGCTTCAATAATATTGAAAAAGGAAGAGTATGAG TATTCAACATTTCCGTGTCGCCCTTATTCCCTTTTTTGCGGCATTTTGCCTTCCT GTTTTGCTCACCCAGAAACGCTGGTGAAAGTAAAAGATGCTGAAGATCAGTTG GGTGCACGAGTGGGTTACATCGAACTGGATCTCAACAG
35	<i>Drosophila</i> EST	LD05707 (AA246767)
	Annotated <i>Drosophila</i> genome genomic segment	AE003727
40	Annotated <i>Drosophila</i> genome Complete gene candidate	CG7444 - very short ORF with EF hand homology
	Human homologue of Complete gene candidate	none
45	Putative function	Possible calcium binding protein

WO 01/72774

PCT/GB01/01297

100

Confirmation by RNAi Slight loss of G1 peak

Example 22 (Category 3)

	Line ID	1439/7
	Category	Mitotic defects in brain: prometaphase arrest.
5		(overcondensation, polyploid, no anaphases, scattered
		chromosomes with bipolar spindles)
	Reversion	?
	Map Position	96F10-14
10	Rescue ID	G3X
	Rescue Sequence	GTCGGATGTAGAAGACGTGCCCCGAAACCCAGTTAGAAATCGATGTCAGCGAT GGCGCCGGACTGGAGGATGAGGATGATGACGATATGGAACAGATTACAGCTC AGAAGGTAAGGTAAATCGTAACAGAGCTTTTTTAATACGCAAGTAATCACATTC 15 TGATATCCCTAGGTTCTGGAAATCATAGAAACCGCGTGGATAAAATGAAATGTG TGCGCCGGAGATCCTGCCCAGCCAGACGGACATGCTGGAGCTGATGGTCTCCC AGGTGGCCCATATGGAGGAGCAGATGCGCGATCTGGACAAGAACGATTTCCG AGCGGTGGTGCACTCCATGGAAGTGGAGAGGGTGGCTACATAATGGCCAGT TATCTGCGTTGCCGCTGCAGAAAGATCGAAACCTTCACGCAGCACATCCTCAA 20 CCAGGAGGAGAGCCGTGAGCCGGATGACAAACGTCTGTCTCCCGAGGAGACT AAGTTCGCCCCAGGAGTTTGCCAGTAAT
	Genomic hit, Accession No.	AC007825
25	Annotated <i>Drosophila</i> genome genomic segment	AE003754
	Annotated <i>Drosophila</i> genome Complete gene candidate	CG14549 – novel
	Human homologue of Complete gene candidate	none
30	Putative function	no homologies which indicate function
	Confirmation by RNAi	Only wild type profile observed

Example 23 (Category 3)

Line ID 1466/4
Category Mitotic defects in brain: metaphase arrest.
(overcondensation, no polyploidy, fewer anaphases, metaphase
5 with bipolar spindle)
Reversion NR
Map Position 72F

Rescue ID ESE

10 **Rescue Sequence 1**

GGCTGGATGCGATTTCGCTTTCGGATTCGGATGGATTCAGCCGCTGTCTCGACA
CCGCCGCAACCGCTCTCGGGAGTTTGAAAATTTGAAATGAGCGGATTTCGCGTT
GCGAAGGCGAGCTAGCGTTGCAGGCAGTGTGGCCAGATGCCGCGTGCGAACG
TATTCTCGAATGCAATCGGCCGAGTGCAGATGCACTAAAAATAACCCACTTCC
15 AGTGACTGGAAATTAAGATCAAGGNAATAGATTTTATAAAAACTTATATGAGT
AAAAATTTTAAAAATTGTGGAGTCAACCTAAATTATAAGCAACTAATTTATAAC
ACAAGTAAAGAATGATATTAAGTAACTTTTAAATAATATTCCATTATGCTTA
CGCTCAATTTATGAACAAATGTTTTCTCGATCCTTAGGTAAAGTTTCGAGTTTC
GCGACTANATTTATTAATAAAGAACATCTCCATTTATGTACACATTTAAAG
20 ATTTATGAGCGGTAATATTAGCTGGTTGAC

Rescue ID ESP

Rescue Sequence 2

ATCCAGCCAAGATATCCTATCGTGCAGCTGAAACCCGAAACCCGAATCCGAGT
25 TCGAAACGAAACGAATCGCAGTGGTGGTTTCTCTCTCGCTCTCTAGCTCTCCCT
CTCTCTCGCGTGTGTGTATGTGTGCGAGTGGCAGGAAAAGTGCGAAGCCGAAA
TCTTTTTAGCTGAAAGAAAGCGCAACTTCAATTAGCGAAAAGCAAGAGTAGCT
AACAAAAAGAAAAGCGGATCGAAAAGTAAAGAAAAACAAAAAAAAAAAAACA
AAAGCAACAAATCGAAATGGCAAGCGAAGTGGCCCAAATACCCGCCGAGGG
30 AAACGCCCGCAGTGGCGGCGGCGGAAAAATCAGAGGAGCCGGAAAAGTCAG
CGGCCCGCCAGCGGACTCAGCGGCCGCTCCAGCTGCCGCCCCCGCAGTGGA
GAAGGCTGAGGATGCCGATGGCGAAAAAAGGACGGCGAGGCCGGAAAGCA
GGACAAGCAGCAGGATGGC

35 **Genomic hit, Accession No.** CSC:AC020154

Associated ORF

Genscan ORF: ORF2 predicted sequences

>21:06:03|GENSCAN_predicted_peptide_5|415_aa

40 MASEVAQIPAEETPAVAAAEKSEEPEKSAAPPADSAAAPAAAPAVEKAEDADGE
KKDGEAGKQDKQDGEPPKKDEAVAAPVATKSEAPPAQKFNVHKTNFEKDIYLY
YQFSRTPLPSLSPYCLKVETWLRVLGLKYENVVDHKMRFRSKKGQLPFIELNGEEI
ADSAMIKELSSKYEKYLDISGLTAEQRNVSYATIAMLENHLIWIIFYWRKYPDNV
LKGYKVNQLHALGLRLPNSILNFFFKITFGRKGTKKLKAHGIGVHSAEEIEEFKGD
45 DLKVLSEMLDCKPFFFGDEPTTLDVVAFAVLSQLHYLSKDIA YPLRDYMTTEKCPN
LIGHVSRMKDKCFPDWDEICTKLDLNAHIPKPEPETKEGKEGGEQKSNEQEGTE

GDKIEKELEKDKSNEKESTEENKEKEETK

>21:06:03|GENSCAN_predicted_CDS_5|1248_bp

atggcaagcgaagtggcccaatacccgccgaggaaacgccgcagtgccggcgccggaaaaatcagaggagccggaaaa
 5 gtcagcggccccgccagcgggactcagcggccgctccagctgccgccccgcagtgaggagaaggctgaggatgccgatggcga
 gaagaaggacggcgaggccggaaagcaggacaagcagcaggatggcgaggagcccaaaaaggacgaggcgggtggcagc
 acccggtggcgaccaaatcggaagccccgcccgcagaaattcaatgtgcacaagaccaacttcgagaaggacatcatctatct
 gtaccagtctcgcgcacccactgctgcccctcctgtgcctactgcctgaagggtggagacctggctgctcttggggcctga
 aatacgagaatgtcgcataagatgctgttccgctccaagaagggtcagctgccgttcacgcagctgaatggggaggaaatcgc
 10 cgattcggccatcatcaaggaactgtcgtccaaatacgagaagtacctggactcgggactaccgccgagcaaaaggaatgt
 ctgtagccacgattgccatgtggagaacctctcatctggatcatcttctactggcgcccaagtatccggacaatgtgctcaa
 gggctacaaggtcaactgcagcacgccctcgccctgcggctgcccaactcgaattctgaacttcttcttaagatcaccttggctgc
 aagggcacgaagaagctgaaggcgcacggcatcgggtgccacagcggcaggagatcgaggagttcgcaaggacgacctg
 aaggtgctcagcgagatgctgcactgaagccttcttcttcggcgacgagcccaccacctggatgtggtggccttcgctgtcct
 15 ctgcagctccactatctgtccaaggacattgcgtatccgctgcgcgactacatgaccgagaagtgcaccaactgattggccacg
 tatctcgatgaaggacaagtgttccccgactgggacgagatctgcacgaagctggacctcaatgcgcacattcccaagccag
 agcccgagaccaaggaggggcaaggagggtggcgagcaggagaataacaaacgaacaggaggggcactgaggggcacaagat
 cgagaaggagttggagaaggacaagtcaaacgagaaggagtcgaccgaggagaacaaagagaaggaggaaacaaagtaa

- 20 **Drosophila Gene Hit** rescue sequence and TBLASTN with ORF2: failed axon
 connections (U21685)
Human Homologue BLASTX with fax: Metaxin 1 and 2 (Q13505 and AF053551)
Drosophila EST several including LD31362 (AA951078 similar by BLASTN to
 U21685 failed axon connections)

25 **Annotated Drosophila genome genomic segment** AE003527

Annotated Drosophila genome Complete gene candidate CG4609 – fax failed axon
 connectionsconnections

- 30 **Human homologue of Complete gene candidate** 4505281
 ref[NP_002446.1|pMTX]
 metaxin>gi|3024205|sp|Q135
 05|MTXN_HUMAN
 METAXIN (4e-06)

35

Putative function Drosophila fax is a dominant genetic enhancer of the Abl mutant,
 developmentally expressed in axons of the CNS

- 40 **Confirmation by RNAi** Weak reduction of G1 and G2/M peaks indicating fewer
 cycling cells

45

Line ID 262/20
Category Mitotic defects in brain: metaphase arrest.
(overcondensation, polyploidy, aneuploidy, few anaphases, high mitotic index, metaphase with bent bipolar spindle)

5 **Reversion** NR
Map Position 72F

Rescue ID G6E
Rescue Sequence

10 AGCTGCACGATAGGATATCTTGGCTGGATGCGATTCGCTTTCGGATTCCGGATG
GATTCAGGAGCCGCTGTCTCGACACCGCCGCAACCGCTCTCGGGAGTTTGAAA
ATTTGAAATGAGCGGATTCGCGTTGCGAAGGCGAGCTAGCGTTGCAGGCAGT
GTGGCCAGATGCCGCGTGCGAACGTATTCTCGAATGCAATCGGCCGAGTGCA
GATGCACTAAAAATAACCCACTTCCAGTGAAGTGGAAATTAAGATCAAGGAAT

15 AGATTTTATAAAAACTTATATGAGTAAAAATTTTAAAAATTGTGGAGTCAACCT
AAATTATAAGCAACTAATTTATAACACAAGTAAAGAATGATATTAAGTAACTT
TTTAAATAATATTCATTATGCTTACGCTCAATTTATGAACAAATGTTTCTCG
ATCCTTAGGTAAAGTTTCGAGTTTCGCGACTAGATTTATTAAAATTAAGAACA
TCTCCATTTATGTTCCC

20

Drosophila EST several including LD28084 (AA949260)

All other results as for line 1466/4

25

- Line ID** 262/22
Category Mitotic defects in brain: metaphase arrest.
(overcondensation, polyploidy, few anaphases, high mitotic index,
metaphase with bent bipolar spindle)
- 5 **Reversion** NR
Map Position 72F
- Rescue ID** F1E
Rescue Sequence 1
- 10 AGCTGCACGATAGGATATCTTGGCTGGATGCGATTTCGCTTTCGGATTTCGGATG
GATTTCAGGAGCCGCTGTCTCGACACCGCCGCAACCGCTCTCGGGAGTTTGAAA
ATTTGAAATGAGCGGATTTCGCGTTGCGAAGGCNAGCTAGCGTTGCAGGCAGT
GTGGCCAGATGCCGCGTGCGAACGTATTCTCGAATGCAATCGGCCGAGTGCA
GATGCACTAAAAATAACCCACTTCCAGTGACTGGAATTAAGATCAAGGAAT
15 AGATTTTATAAAAACTTATATGAGTAAAAATTTAAAATTGTGGAGTCAACCT
AAATTATAAGCAACTAATTATACACAAGTAAAGAATGATATTAAGTAACTT
TTTAAATAATATTCCATTATGCTTACGCTCAATTTATGAACAAATGTTTTCTCG
ATCCTTAGGTAAAGTTTCGAGTTTCGCGACTAGATTTATTAATAAATAAGAACA
TCTCCATTTATG
- 20 **Rescue ID** F1P
Rescue Sequence 2
- 25 GTGCAGCTGAAACCCGAAACCCGAATCCGAGTTCGAAACGAAACGAATCGCA
GTGGTGGTTTCTCTCTCGCTCTCTAGCTCTCCCTCTCTCTCGCGTGTGTGTATGT
GTGCGAGTGGCAGGAAAAGTGCGAAGCCGAAATCTTTTAGCTGAAAGAAAG
CGCAACTTCAATTAGCGAAAAGCAAGAGTAGCTAACAAAAAGAAAAGCGGAT
CGAAAAGTAGAGAAAAACGAAAAAAAAAAAAACCAAGCAACAAATCGAAATG
GCAAGCGAAGTGGCCCAAATACCCGCCGATGAAACGCCCGCAGTGGCGGCGG
CGGGAAAAATCAGAAGAGCCGAAAATCAGCGGGCCCGCCAGCGGGACTCTG
30 CGGGCGCTCCAGCTGCCGCCCCCGCAGTGGAGAAGGCTGAGGATGCCGATGG
CGAA
- Drosophila EST** several including LD28084 (AA949260), LD38479 (AI518768)
- 35 Other results as for line 1466/4

Line ID	262/3
Category	Mitotic defects in brain: Metaphase arrest (overcondensation, polyploidy, aneuploidy, no anaphases, high mitotic index, metaphase with bipolar spindle)
5 Reversion	NR
Map Position	72F
Rescue ID	H3E
Rescue Sequence	
10	AGCTGCACGATAGGATATCTTGGCTGGATGCGATTCGCTTTCGGATTCGGATG GATTCAGGAGCCGCTGTCTCGACACCGCCGCAACCGCTCTCGGGAGTTTGAAA ATTTGAAATGAGCGGATTCGCGTTGCGAAGGCGAGCTATCGTTGCAGGCAGTG TGGCCAGATGCCGCGTGCGAACGTATTCTCGAATGCAATCGGCCGAGTGCGAG ATGCACTAAAAATAACCCACTTCCAGTGACTGGAAATTAAGATCAAGGAATA
15	GATTTTATAAAAACTTATATGAGTAAAAATTTTAAAAATTGTGGAGTCAACCTA AATTATAAGCAACTAATTTATAACACAAGTAAAGAATGATATTAAGTAACTTT TTAAATAATATTCATTATGCTTACGCTCAATTTATGAACAAATGTTTTCTCGA TCCTTAGGTTAAGTTTCGAGTTTCGCGACTAGATTTATTAAATTAAGAACATC TCCCTTTATGTTC
20	Other results as for line 1466/4

Example 24 (Category 3)

Line ID	238/20
Category	Mitotic defects in brain: metaphase arrest (overcondensation, metaphase with bipolar spindle)
Reversion	NR
Map Position	75E1-3
Rescue ID	D7E
Rescue Sequence	TTCAGTCGCGCATTTCACCGTTTCCGAATCGGACGAACCGGGCGTGATTGCTC TCCTGCTGCTTTTCGAGATCGGAGTCCCGATAAGGATATAACTACAACCTAAAG AGGAATCCAAGCCTCCTCCTGCCGCTAGTTTTCGAAAAGTAAATAGAGTACTTG TTATCAACTGGGGAAGCGGAGATACATAGCTCCGATATTCTGTGAAAGCCAG ACAAACGGGATACCAACGAACAATCGCCATGTGCGTCGTCCCTTCTCGTTT CACACATCGTGCGATAAAAAATACCGCTTTTGCTTTTTGTGTTTATTTAAAAATTT TGGTTAGGAAGTGAACCTCGAACTCGTGACGTTTGCATTTTCACAACAACAAAA AGAGCAAAACATAGCAGAAGAACCCCGAAGAAACAGGAACAGAAACCGTT GACCGAGTGCCAGTGTGAAGGTCTAGGCACAAAGAACGCTACCAAGAACTCT TGGGAGTTAGGGAGGCTCTTTACAATGACAACATTGCACCAAGATGGACTCT CTCTCTAAAATGCATTTTCATACCAATATTACTTT
<i>Drosophila</i> EST several including LP04802 (AI260815)	
Annotated <i>Drosophila</i> genome genomic segment	AE003519
Annotated <i>Drosophila</i> genome Complete gene candidate	CG3979 - novel gene with homology to sodium- dependent dicarboxylate transporters
Human homologue of Complete gene candidate	3e-87 4506979 ref NP_003975.1 pSLC13A2 UNKNOWN >gi 2499523 sp Q13183 NDC1 _HUMAN RENAL SODIUM/DICARBOXY
Putative function	sodium/dicarboxylate transporter
Confirmation by RNAi	Only WT profiles observed

Line ID 490/9
Category Meiotic defects in testis: segregation defects, multipolar spindles (Mul-02/29)
Reversion NR
5 **Map Position** 95C1-8

Rescue ID I4E

Rescue Sequence

GCTCTGCCGCTTCAACCGCCCGCGTTCTGTGTGTTGGTGTGCCGCGACGTAGG
10 TGTAGGGTCCGCTGCACACGTGTGTGTGGGAGCGCGCGAGAGCGGGAGAAGA
GCAGAACGTTTTTGGGCGGCTAGTGGTGGCACC GTGAGCATGCCGGTCGTCGT
AAGATAGGCTTAGGAACACTCAGAGAAAATTTGTTAGCTCAGCATTTTCCTA
TTATTGAAATCATTTATTTGATGGTCTATGGGGGTTTCTTCGTAGTTATTCAT
AGATCGGCGATTTAAGCTACGCTTAAAGGGTAATTTGTCTGAGATATCTTTGT
15 CATTTAAAGTTAAGTCTCAGCTTATCCAAAAGTCAGTTATTGGAAAAAAGGAG
CCAGCTTTTCAGCAGAGTTCGGCTTAAAGCGCTTATTATCATATTAACCAGCTTA
ATTAATGTATCTTTTAAATTGTTATATGCATTAAATCACTAATTAAGGTGATTA
CCATTTGTACGTTTTTAAATTAAAGTATTTTGATTTTCACTAATACAGGCTCTAA
GCTGATCCAAATCTACAAGCTTAGTTTTTGAATAGTCTTCACATGTTGACTTTT
20 ATTCTCT

Genomic hit, Accession No. CSC:AC015160

Other results same as 238/20

25

Line ID 660/3
Category Meiotic defects in testis: cytokinesis defects, abnormal spindles.
(Ab-01/03)
Reversion R?
5 **Map Position** 75E

Rescue ID H8E
Rescue Sequence
GCTCTGCCGCTTCAACCGCCCGCGTTCTGTGTGTTGGTGTGCCGCGACGTAGG
10 TGTAGGGTCCGCTGCACACGTGTGTGTGGGAGCGCGAGAGCGGGAGAAGA
GCAGAACGTTTTTGGGCGGCTAGTGGTGGCACCGTGAGCATGCCGGTTCGTCGT
AAGATAGGCTTAGGAACACTCAGAGAAAATTTGTTTAGCTCAGCATTTCCTA
TTATTGAAATCATTTATTTGATGGTCTATGGGGGTTTCTTTCGTAGTTATTCAT
AGATCGGCGATTTAAGCTACGCTTAAAGGGTAATTTGTCTGAAATATCTTTGT
15 CATTTAAAGTTAAGTCTCAGCTTATCCAAAAGTCAGTTATTGGAAAAAAGGAG
CCAGCTTTTCAGCAGAGTTCGGCTTAAGCGCTTATTATCATATTAACCAGCTTA
ATTAATGTATCTTTTAAATTGTTATATGCATTAAATCACTAATTAAGGTGATTA
CCATTTGTTTCGTTTTAAATTAAAGTATTTGAATTC

20 **Genomic hit, Accession No.** CSC:AC015160

Other results same as 238/20

Example 25 (Category 3)

5	Line ID 273/18 Category Mitotic defects in brain: metaphase arrest (overcondensation, very high mitotic index, few polyploids, metaphase with bipolar spindle) Reversion NR Map Position 75E
---	--

10	Rescue ID D1E Rescue Sequence AACTGGGCTAAAACCAGCTGAAACTGGTGAAAAGTAAAATATTTGGAGAAG GAAAGCCTTAAGTTCCTCTCTACGCTTCGTACACGTAATGTGCGTGGTTTAATC TACGTTAAAACAAGTGGAACCATGTTACGTGCCGTGGCTTTGTGTGTGTCAG TGGTGCTCATAGCACTATATACGCCAACTTCTGGGGAATCCAGTCAGAGCTAT CCCATTACCACGCTAATCAACGCGAAATGGACGCAGACGCCCCTATATCTGGA AATCGCCGAGTATCTGGCCGATGAGCAGGCGGGCCTCTTCTGGGATTACGTTT CGGGGGTGACAAAGTTGGACACGGTTCTCAACGAATATGGTTTGTGTTTATAA GTCATGGAGAACCCGCATTAAAGAGCTTTTATATTCTCCTCAATGTGAATCC GAATCCATATAAAATC
----	---

Genomic hit, Accession No. AC015160
Associated ORF
 Genscan: >ORF2 predicted sequences
 >16:57:34|GENSCAN_predicted_peptide_5|1548_aa

25	MLRAVALCVSVVLIALYTPTSGESSQSYPTTLINAKWTQTPLYLEIAEYLADEQA GLFWDYVSGVTKLDTVLNEYDTESSQYNAALELVKSHVSSPQLPLRLVSMHS LTPRIQTHFQLAEELRSSGSCQSFTFAQVGSELACSFNELQKKLEVPLAKDSLDA VVTYSFDHIFPGSENNRTRTVVLYGDLGSSQFRTYHKLLEKEANAGRIRYILRHQLA KKDKRPVRLSGYGVELHLKSTEYKSQDDAPKPEAGSTSDEDLANESDVQGFDFK VLKQKHPTLKRALDQLRQRLQGNDEIAQLKAWFQDLGLQAAAAIAEIQGD LQILQYTAHNFPMLARTLLAHKVTDGLRAEVKHNTAFGRSLNVAPPDGFALFING LFFDADTMDLYSLIETLRSEMRVLESLSNNVRGSLASSLLALDLTASSKKEFAIDI RDTAVQWVNDIENDVQYRRWPSSVMDLLRPTFGMLRNIRKNVFNLVLVVDAL QPTARSVIKLSSEFVIHQAPIRLGLVFDARDANEDNLADYVAITCA YNYVSQKKD ARAALSFLTIDYAAVGETKVVTKKDIVKQLTKEFTSLSFAKAEFLEEDSTYDYGR ELAAEFIQRLGFGDKGQPQALLNGVPMPSNVVTADSDFEAAIFTEIMTHTSNLQKA VYKGELTDNDVAIDYLMNQPHVMPRLNQRLSQEDVKYLDINGVAYKNLGNVG VLNRLSNRDMTATLMDNLKYFGGKKSTELIGRASLQFLTIVWFADLETQGRDLL THALDYVQSGESVRVAFIPNTESSASSRRNLNRLVWAAMQSLPPTQATEQVLK WLKKPKKEIPTQLEDILGSTELHLKMLRVYSQSVLGLNKSQRLVIGNGRLYGPL SSDESFDSDALLARFSSLQYSDKVRQVLKESAQDVNEEFNSDTLLKLYASLLPR QTKTRFKLPTDLKTDHSVVKLPPKQENLPHFDVAAVLDPASRAAQKLTPILILLRQ VLNCQLNLYLIPVPQHSDMPVKNFYRYVVEPEVQFEANGGRSDGPLAKFSGLPAN PLLTQQLQVPENWLVEAVRAVYDLNKLTDIGGPVHSEFDLEYLLLEGHCDFDAA SGAPPRGLQLVLGTQSQPTLVDTIVMANLG YFQLKANPGAWSLRLREGKSADIYA
----	---

10 >16:57:34|GENSCAN_predicted_CDS_514647_bpattgttacgtccgtggcgtttgtgtgtctgtgtgtctca
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 ccaaactcacggccgcccagaggattgtcccgaatggaaggactacgatgccgagctgaagaccctgatgtctgcacatcag
 15 gatcatgagaattcgcatagcagggactcggcagttgatgattcgggtgacgattcgggtgaggtcaccactgtgacgccttctcat
 gagcccaagcacggcgagctgtga

***Drosophila* Gene Hit** rescue sequence and BLASTX with EST and TBLASTN with
 ORF2: UDP-glucose:glycoprotein glucosyltransferase (U20554)
 20 **Human Homologue** BLASTX with UDP-GGT: hypothetical protein (AL133051)
***Drosophila* EST** several including GH16576 (AI293351)

25 **Annotated *Drosophila* genome genomic segment** AE003519
Annotated *Drosophila* genome Complete gene candidate ugtUDP-glucose-glycoprotein
 glucosyltransferase

30 **Human homologue of Complete gene candidate** CG6850-
 IGI_M1_ctg14521_41
 D65BCE6EEC187AE3
 TRANS:SEPT20T.ctg14521.2
 2 FPC_ctg:ctg14521
 FPC_start:1284609
 FPC_end:1284696
 35 FPC_strand:+ (1.20E-215)

Putative function ugtUDP-glucose-glycoprotein glucosyltransferase

40 **Confirmation by RNAi** Only wild type profiles observed

Example 26 (Category 3)

5	Line ID 430/5 Category Mitotic defects in brain: metaphase arrest (overcondensation, polyploidy, metaphase with bipolar spindle) Reversion NR Map Position 98B5-8
10	Rescue ID 2C2E Rescue Sequence GTGCGGCCCATGGATGTGCGAACGTGTACGAAGACCAAGATCGGCATCGCCA TCGGCGGCAGCACGACGGACGATAACGAAAAAGCTACAGCCGCCGCCACAGA TACAGATGCAGATGCCATGCCGCTGTTATCAGCGCGAGCGGGAGAATGATAA GGGATGGGATCGCTCAGCGCGGCAGGCAAGACGACCAAAAAAGAGAGCCAAC 15 TAAATGATGTGCCTAAGACTAAGAGTTTAATGAGCATTACTGTCGCGCACTCT ATGTATTATGAATAAAATTCATACAACCTTTGTGGTTTATTATAATAAAAAGT GTGTCAGCTCTACTCGGGGGAAAGTAAGTTTACTTCTTGGCCGCTGGCTTCTTG GCGGCGACCTTCTTCTTGGCGGCGGCCAGCAACTTGGCGCGATTGGCGCAGCC TTGGTGGCCACATTGGCGAAGTGCGACTTGGCCAGCTCGACGTTCTGCTTCTT 20 GGCTTGGCCAGCACCTTGGCCACGGTGCGCTTCTCGGCGGCGAGGGCGGCAC GACGCTTGAGTACCTCGGCATAAGGGTTCAACTTGATCAACTTGCACGCGTT GGTAAGGGGGTT
25	Drosophila EST several including LD45359 (AI513164) Annotated <i>Drosophila</i> genome genomic segment AE003763 Annotated <i>Drosophila</i> genome Complete gene candidate CG5502 RpL1 - Ribosomal protein L1
30	Human homologue of Complete gene candidate 1e-126 432359 dbj BAA04887 (D23660) ribosomal protein [Homo sapiens]
35	Putative function structural protein of ribosome involved in protein biosynthesis
40	Confirmation by RNAi Marked decrease in G1 and G2/M indicating fewer cycling cells

Example 27 (Category 3)

	Line ID	472/12
	Category	Mitotic defects in brain: metaphase arrest. Meiotic defects in testis: segregation defects. Abnormal spindles (mitotic: High mitotic index, meiotic: Ab-08/24)
5	Reversion	R?
	Map Position	96C7-9
	Rescue ID	2B6E
10	Rescue Sequence 1	GTCTGACGTTCTCTGAGGGCAAAAGTTTCGAGTTAGTTGAAGGTGAGGGTGCT CGATCACCGATTGCGGTGAGACGAAAGAAAAGTATGCATTGTTGCGTTGTAA AGAGAGCCGCGCTCGTCTTGTTTCACATTGTCGCTGAGAACGTATGTTGTGCT TCATCATTTCCCTTGTTGATTTCCCTTTGACGTGGCAACTTGACCATGTATGACA 15 ACTCTTTGGTGGTGCCATCTGGAAGGCAGAAATTTGATGTCAACGGTGCTCCC AGCCAGTCCACTCCCCAACTCACCTGCAGCTCCACTTCGATATTAACGCGCA ACATATTAGTGGCGTAGTTGTCACCTGCCGCGGATCCCATTTCCGCTTTGAAAT TTCGCACTTTCGAATATCCGTCCACATTCGATTTGAGAACATCTTCGAAACGTT CAGCGGTGACCCAATCGGGTATTTTGCCAGCCGCCATTGTAGATAATCGGGAT 20 AAGTATTTTGAAATCGAGCAGAAAACACATATACGTCCAGTGTGACGGTCTTG CGTAGACTGATGAAAGCCGAGTATTAGACTCTACACATCTGTGGAGCTTTTA ATTCGTAGTGCGCGGCCGATTCTCTCGATCTTCTCTCAAAGCTCCGCTAAT
	Annotated <i>Drosophila</i> genome genomic segment	AE003751
25	Annotated <i>Drosophila</i> genome Complete gene candidate	CG10618 - novel
	Human homologue of Complete gene candidate	none
	Putative function	no homologies which indicate function
30	Confirmation by RNAi	Only wild type profiles observed

Example 28 (Category 3)

5	Line ID	571/15
	Category	Mitotic defects in brain: metaphase arrest (overcondensation, few anaphases, some polyploids)
	Reversion	NR
	Map Position	93D
10	Rescue ID	2A8E
	Rescue Sequence	GGCGGCGCTACATTTGTTGTTGTCGCTGCTGCTCACAGCTCCACCACCATTTGC ACAGTTATATTACCTCGCTCAAGTCGCCCCCTCTCCCTCTCGCCCACTCGCTGTG TCAATCGAATTAAAACGAATGCTCTTCGGCGAATAATTGGGTTTAGATACTTT TCCAGCAGACAAAAGTTGTATTTTTGCACTTCTTATTGATATTAGGCAAAACGC 15 ATCGGCCGAATCACACGCACACAAAGCACACACGCGAGCAGCGGTTTTCAA TCTGCAGTACACCAAACAACACACACTATTTCTAATGCCTGTTCTTATCCCTC TGATATTCCCAATGAATCGCTGGGCAATTGGCGATTCTGAACCGATTTTCACTT GGCTCTTTGTTTTATTTAATTTTACCGAAACGCTCTCACACGCAGAGACGCTT TTGCTCGTTCGCTGATGCTTCTGCTGCAATACACACCACCTACGAAACGAGCC 20 AAGGGAAATTGTATCTATGGGCTGTGTATCTGTTTCTACGCGGCACGCGCTGC ACGTCCGCTCGCTTCGGGTTTTCGAGAGAGAATATACTTTTTCGATACGGTA CGGTAAACGAATTCCGCGGAATTAATTCTTGAAGACGAAAGGGCCTCGTGATA CGCCTATTTTATAGGGTAATGCATGATAATAATGGGTTCTTAGACGTCA
25	<i>Drosophila</i> EST	LP07504 (AI294185), LP06548 (AI293427)
30	Annotated <i>Drosophila</i> genome genomic segment	AE003734
	Annotated <i>Drosophila</i> genome Complete gene candidate	CG15802 – novel homology to Doom, a product of the <i>Drosophila</i> mod(mdg4) gene, induces apoptosis and binds to baculovirus inhibitor-of- apoptosis proteins
35	Human homologue of Complete gene candidate	none
	Putative function	inducer of apoptosis
	Confirmation by RNAi	Only wild type profiles observed

Example 29 (Category 3)

	Line ID	736/15
	Category	Mitotic defects in brain: prometaphase arrest (overcondensation, fewer anaphases, metaphase with bipolar
5	spindle)	
	Reversion	NR
	Map Position	73C
	Rescue ID	H5E
10	Rescue Sequence	
	CTAATGAGTAAGGAAAACCAATCAGCCTTGCTAATCGCTTGGCAGTATTGGCT TCTATGCAGGGGGGCGTGTCCCGCGCCCTTGAAGCTCAAATTTTTCGCAAGGG CACAGGTCGTCCCTCCTCCTCCGCGTGGGTGGCGTTCGGCCGAACGAACCGG CGCCTACTTTGCGTCCGGCTAGCGAGGATCTCTGGGTGCCACCCACGGCTGG GTGTTGCGATCTGCCCGATTGATAATCCATGCGTGAGAAAGCTTTAGAGAATC TGCCAGATTATTACTCCCCGCATACTCAGAAAAATGTATCCTTCAGATATG TTTATGTTTATGAAGTGAAAAAAGTCCTTTGAAATACTACAAAAAGTGAGGAT CTGACCAATGATTTGATTTCTATAGAAATATACTATAAACTATAAACTAC	
20	Genomic hit, Accession No.	CSC:AC014181
	Annotated <i>Drosophila</i> genome genomic segment	AE003526
	Annotated <i>Drosophila</i> genome Complete gene candidate	CG3971 baldspot - with homology to membrane glycoprotein
25		
	Human homologue of Complete gene candidate	CG3791-9e-08 4680391emb CAB41293.1 (AL034374) dJ483K16.1 (novel protein) [Homo sapiens]
30		
	Putative function	membrane protein, function unknown
35	Confirmation by RNAi	Slight reduction of G1 and G2/M peaks indicating fewer cycling cells

Example 30 (Category 3)

	Line ID	82/24
	Category	Mitotic defects in brain: metaphase arrest (condensation, no polyploidy, no anaphases, metaphase with bipolar spindle)
5		
	Reversion	NR
	Map Position	100D
10	Rescue ID	2E3E
	Rescue Sequence	GGTCAAGCCCCGATGGCGTCCAGCGCGGGCTCGTCGGCAAGATCATCGAGCGC TTCGAGCAGAAGGGCTTCAAGCTGGTCGCCCTGAAGTTCACCTGGGTAAGCGG ATAATTGAATTAGGAAGAAATCAATAGATATACATACGTGGAAACGGGTTGC 15 CCCACGCGGGGTTGCTATCGGACCTAACCTCAAAGGCTGGGTGCAGGCGTCAT CGCGGAATGACATGTGTTTAGAGGTCAGAACTGCAATTAAGTATAACGAACC GTTTTGTAACCAGGCCTCCAAGGAGCTGCTGGAGAAGCACTACGCTGATCTGT CCGCCCCGCCCTTCTTCCCCGGACTCGTGAAGTACATGAAGTCCGGCCCCGTG GTGCCCCATGGTGTGGGAGGGTCTGAATGTGGTCAAGACCGGTCGCCAGATGCT 20 CGGCGCCACCAACCCCGCCGACTCGCTGCCCGGCACCATCCGCGGTGACTTCT GCATTCAGGTCGGACGCAACATCATCCACGGCTCCGATGCCGTCGAGTCTGCC GAGAAGGAGATCGCCTGTGGTTCAACGAAAAGGAGCTGGTCACCTGGACCCC GG
25	Genomic hit, Accession No.	CSC:AC012727
	Associated ORF	Genscan ORF1 predicted sequences >16:43:49 GENSCAN_predicted_peptide_7 172_aa MKLLMLGTLAFFSVISATMAANKERTFIMVKPDGVQRGLVGKIIERFEQKGFKLV 30 ALKFTWASKELLEKHYADLSARFFPGLVNYMNSGPVPMVWEGLVNVKTGRQ MLGATNPADSLPGTIRGDFCIQVGRNIHGSDAVESAKEIALWFNEKELVTWTPA AKDWIYE
	>16:43:49 GENSCAN_predicted_CDS_7 519_bp	atgaagctcctgatgctcggcacaaatttggcattctttctgtaatctcggcgacaatggcggctaacaaggagaggactttcatcat 35 ggtcaagcccgatggcgtccagcgcggtctgctggcaagatcatcgagcgcttcgagcagaagggttcaagctggctgccc tgaagttcacctgggctccaaggagctgctggagaagcactacgctgatctgtccgccgcccccttctccccggactcgtgaa ctacatgaactcggccccgtggtgccccatggtgtgggagggctgaatgtgtcaagaccggcgcagatgctcggcgccac caaccccgccgactcgtgccccggcaccatccgcggtgacttctgcattcaggtcggacgcaacatcatccacggctccgatgc 40 cgtcgagtctgccgagaaggagatcgccctgtggttcaacgaaaaggagctggtcacctggaccccgccgccaaggactgg atctacgaatag
	Drosophila Gene Hit	rescue sequence and TBLA: abnormal wing disc (awd) (X13107)
45	Human Homologue	BLASTX with awd and TBLASTN with ORF1: tumor metastasis inhibitor nm23-H2 (A49798) non-metastatic cells 2, protein (NM23B) (P22392) and nucleoside diphosphate kinase B.

***Drosophila* EST** several including LP05977 (AI257573 similar by TBLASTX to X92956 B.taurus mRNA for nucleoside diphosphate kinase (NBR-A)

- 5 **Annotated *Drosophila* genome genomic segment** AE003779
 Annotated *Drosophila* genome Complete gene candidate CG2210 - awd abnormal
 wing discs nucleoside
 diphosphate kinase
- 10 **Human homologue of Complete gene candidate** gi4505409
 1A5C3F84D7AD272C
 [ref]NP_002503.1| non-
 metastatic cells 2, protein
 (NM23B) expressed in [Homo
15 sapiens] (1.90E-61)
- Putative function** human nucleoside diphosphate kinase, transcriptional regulation of
 c-myc expression.a candidate suppressor of tumor metastasis
- 20 **Confirmation by RNAi** Only wild type profiles observed

CATEGORY 4: ANAPHASE DEFECT**Example 31 (Category 4)**

	Line ID	1132/8
5	Category	Mitotic defects in brain: anaphase defects (overcondensation, high polyploidy, some lagging chromosomes)
	Reversion	?
	Map Position	86F3-6
10	Rescue ID	2C3E
	Rescue Sequence	
	GGCCGGAGGTACCATTTTGGTAGGACCGTTTTTCGGGCCAACGAAAATACCAC	
	AAGACGGCAGCGATAATAGTGTTTTTTGCTTCAAATGTAGTATGGCTACGCAA	
	CTCACATATGGTTAAGAACTTCGCTGTTTATTTGGTGGTTAACTAGCTAAATA	
15	CAATAAGAGTGGCAACGCCGTCACGTTTTCTACATGTATTTTACTTGGCGTAGT	
	GCGCCAAGCTTATAAACCACAGTTGGGCGGTTCTTTTGAATTGTTAATTTACA	
	CCCCACTATGAACTTATTAGCCTTCTTTATTTATTTTATATTTTATTTTATTTT	
	AGAAATACGTTTACTCAAGGTTTCGCAGCTTGTCAATCAGTATTCGCAAATATCA	
	ATAATAAAAGGCATCAATTTTCCAATCAGCAGTTGAAAAGAACTCCCTCGAC	
20	ATTTGAACAAAATGCATTTTTGGGTGATTATAATTTATTAGAATTTTTATTGAC	
	TTAAGGTAAATATAAATAAAATATTATTCAAGTACAAAGGTATATATACTCAT	
	TAATANTATTTGGATTCAAGGAAAATATATTCAAATGGCGGGGGTTTAATA	
	AAACAATTTTTCAAATTAAGG	
25	Genomic hit, Accession No. AC007805	
	Drosophila EST	several ESTs including LP09688 (AI295922)
	Annotated Drosophila genome genomic segment	
30	Annotated Drosophila genome Complete gene candidate	AE003693 CG6929 - Lk6 kinase
	Human homologue of Complete gene candidate	
35		
40		

Putative function Protein kinase associated with microtubules

Confirmation by RNAi
cells

Complete loss of G1 and G2/M indicating fewer cycling

Line ID 483/19
Category Meiotic defects in testis: segregation defects
Reversion ?
Map Position 86F

5 Rescue ID H2S
Rescue Sequence 1
CTCCGGCCACACGGATGAATTCGTCGTCATTCGTCGGAATCATTCGAACTTTG
AAAATGGATCGGTAGCTGGGAAGGAACTTAAAGCGAAATACGCAAAGAAA
10 ACGGCTTTTGTCCGCTATTCAGCGATTTTTTTTGTGTTGTAATCAGCAGAGGAA
ATTTTAACGACCAACTCCACCGCCACACCAGCCATCTCCAGCAGCCCCGGAAA
ATAAAATAGAACTAAATTAACGCCACCATCACTACAACAACCATCTCACCAAC
AACTACAAGAGCAACAACCACAGCAACAGCACTACTGCACCAAGCCCACAAA
GAAGAGGTGAAACGCAATAATCGA=CAATACCCGAAGAAAAAACAACAAAA
15 ATATCGCAGATAACCGAAAAAAGCGGTGCAATAGATAAACCCCATTTTTTGCT
TGAGCTTTTTTCGCCTGTGTGATGAGAGAAATCAGCAGCAGCCATCGATTACA
ACAACAACAGCAGCCACACCAACGACGACTCACCACCAACGAAGAATAATA
ACCAGCGGANAGCGATAGATA

20 Genomic hit, Accession No. CSC:AC018284
Drosophila EST several including GH28825 (AI517767), LP04213

Other results same as 1132/8

Example 32 (Category 4)

5	Line ID	1422/14
	Category	Male and female sterile, small wings, meiotic defects in testis: segregation defects, elongation defect
	Reversion	NR
	Map Position	90B4-8
10	Rescue ID	2F1E
	Rescue Sequence	GGCCAGCTGCTCAAACATTCTGCAGCTATTTGGCCGCCAGCGAGTAGAACGAT ATTGCCAAATATTTTATAATAGTAACCAATACGTTACCAGTATGACCGCGCCG ATAACGATAGAAAATACCACACGGTCTAAAAGTAAATACCATTGGGGTATTC 15 CCTAATCTTTTGAATTATTTACCGTTAGGTTTCGGTCTGTTTTTTTTTGTGCTGCTG TTCTTTGTATGAAACGGATTAGTAATTTTATTTGTTGTTTTTGTGCATTTTTTGA TATTAAGCCTTGAAACATGCCTTAAATCGTTAAAATAGATTATAAGAGGGA TGGACTGTTTGTTAAAACCAATTGAAAATTTGTAATCGCTGGTAATAACTAT CGAGATAAGCTTAATTATCGCTGTTTTCTTTGTATCTAGTTATAAATAATAATA 20 ATAAACTGGTAATTAACAAAAGTAAAAAGTTACTTAAGTTATACAAAAATAT TTAGTTATTGNATTCAATAATAAGATGGTAATAATAGATGGTAAGATAGTAAT ATTTTAATAATTGAATTTTCATCACACATGCTGGTGCACGTTCCACAAGTTACAA TCAAACGAAA
25	Annotated <i>Drosophila</i> genome genomic segment	AE003718
	Annotated <i>Drosophila</i> genome Complete gene candidate	CG7623 - novel with homology to UDP-galactose transporter.
30	Human homologue of Complete gene candidate	2136348 UDP-galactose transporter related isozyme 3 - human >gi 1669564 dbj BAA13527 (1e-36)
35	Putative function	sugar modification protein

Confirmation by RNAi **Slightly reduced G2/M**

Example 33 (Category 4)

	Line ID	1479/10
	Category	Mitotic defects in brain: anaphase defects (overcondensation, anaphase bridge, metaphase with swollen chromosomes and bipolar spindle)
5	Reversion	NR
	Map Position	69F3-7
	Rescue ID	2D6E
10	Rescue Sequence 1	CCACGGGCAAATGTGGTCCGGAGGTCCACGACAACGTGCCGCTGACCATATC CCAGATTGAGCGCGCAACTCAGGATCCGGAGAACGAGAATGTGTTTCATCACA GACGACGTGCATCCGATTCACTTCTGCACCTGCATCATCTACGCCTTTGTA GGCAATGGAACGCACAACGAGTCGTTTCATGAAGTTCATGATCGATGATGGCA 15 CCGGCTCCCTGGAGGCCAGCATCACAAAAACCCTTCAATGGACGCGTGATC AGCAGCCTGTACAGTGAAGCCAGTTCGCTGGCCTCGTCCGAGGCCTACAAGA GCATTGCCGTGAGCATGATGCGGCTGCTGCAGGTCTCCATGGAGTACATTGAT CCCACGCGCATCTCGAGGGGCCACAGCCTATTCCTGCGCGGTCGTCCGAATAG GTTCCGCGGCAAGATGGGTCTGGACGCTTTTCAGTTCTTCATAGACAGCGGCC 20 GATCGCGGAATATGGAAATTGGCTTCGTGGACTACCTAACCGACTGGCAACG AAGGCATAAAACAATGCAAAATAC
	Rescue ID	2D6P
25	Rescue Sequence 2	GCCCCGTGGACTTTTCACTCTGTTGATTCTTGCGTATCACGAAGTTATCCAGCTG GCTTTCTATGTCCTCGAACTCTGATTAAAATCCATTCTATTTGCTTAGTCTGC GATTTCAAAGGGGATTTCTTTATTGCAGTGCATTTTGCATTAGCGCCAAAAAA AAAAAAGTTGTGAGCATGGGCGTAGACTTCGTATTTTCTTACAAATAATATTA ATTAATAATTAATTTTGTGAGCAATTTTACACAATTGTATTATAAGTTAAAACC 30 AGGGTCACATTAATTTGCAGAACCGCGCAATATTTTCTTTTAACCCCTTACA AATTTTCAGTTGTTTGTGACTACGCCCTGCTAATTTTACTTATTAAATTCAA GTCTAAAAACATTGTCACCAGATAATACGAGTATACACTATATGGACAAACGT AAAATCGTTAATAGAAATATATATTCAACCATTATTTACCACCGAGAGAAA TTCATTTGCACAAAACGCCAGGTTGGCAGCACCATCATTGCGCACAGCAAGTG 35 GGCAAACTCGTTGTATCGCTTG
	Genomic hit, Accession No. AC007328	
	Associated ORF	
40	Genscan ORF1 predicted sequences >17:42:01 GENSCAN_predicted_peptide_2 1507_aa MKLAPTVKLNNGYEMPILGLGTYNLKKSRCEAAVCHALEMGYRHIDTAYLYRNE GHGKVLAKLIGDQKLKREQVFLVTKLWDIYHEPKMVKYACDMQLKLLGVDYID LYLMHSPVGVDYISDEDLMPHENGQLRTNDVDYVDITYRSMEQLVHLGLVRSLG LSNFFANQLKRLLENCQIKPANLQIECHPELVQVPLIELCKFHNTTVVAYSPLGRSQ 45 TCNPLPDYYTDSKLLALAAKYGKTPAQILRYLSKDNEGEAAVKHAIDVGYRHID TAYFYQNEAEVGKAIRDKIAEGVVKREDIFLVTKLWNIFHDPERVEGICRKQLSNF GLDYIDL YLMHMPVGKYVDDNTLLPKNEDDVLQLSDVDYLD TYKAMEKLVKL	

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 gaagcctgcagctccgagcatctgtcagaattcgacaacgccgggcaacaaccgcagatactcctacagccaaatcagcaatg
 30 tgcgtgactgcgagaaaaaggaggggccatggagtggatgga

Drosophila Gene Hit BLASTN with rescue sequence 2: Histone acetyltransferase GCN5
 (AF029776) very small match at end, TBLASTN with ORF1:
 middle domain histone acetyltransferase GCN5 (AF029776).
 Genomic matches histone acetyltransferase

Annotated Drosophila genome genomic segment AE003541
Annotated Drosophila genome Complete gene candidate CG4107 -Pcaf /GCN5 histone
 acetyl transferase
 transcriptional activator
 protein
 gi6382076
 72F516F8BD10CD0C
 [refNP_003875.2] p300/CBP-
 associated factor [Homo
 sapiens] (1.20E-197)

Putative function Transcriptional activator

Confirmation in RNAi Only wild type profiles observed

Example 34 (Category 4)

	Line ID	184/5
	Category	Mitotic defects in brain: Anaphase defects. (overcondensation, aneuploidy, some lagging chromosomes and breaks)
5	Reversion	R
	Map Position	71B
10	Rescue ID	C4E
	Rescue Sequence	CTCGAGCAGATGTGGGACGAGCTGAGCGGAGCGCACAACTGCCAAGTAAGT GGAGCATGTGGATGAAAGGAGTTCCCAGAACAGTGTTGCCAACCAAAAAAAAA AAAAAAGTTAAAAAGTTAATTTTAATAGTGTAATAAATATGAATTAAATTAA 15 ATTTTTATGTAAACAGTATTAGCTTTACATGAGATTACCAAATTGTGAGTGTCT GTGTTTGTGTTGTCTTTTAAAAACTTTAAAAGCACATAAAGAAATATATTTTAAA TTTAATTAAAAAGTTCGTAAAAAGTAAAAGGTAGCTAAATTAAAAAGTTTCCT ATTCAAATCAGATTTGGCGAACAAAGAGCCAAGTTGGCAACACTGACAATGA CTCCAAGCGCGAACAAAGCGATTTCTATCGTTATCCCACTCTCTCTCCAGAG 20 ATCGTTCTCAAGGCCAAATGGAAGGGACTTCGAGACAATTTCCGTGTGGAGTC AAAAGGATCCGGCGGCCGAATAACGG
	Genomic hit, Accession No.	CSC:AC019852
25	Associated ORF	Genscan ORF1 predicted sequences >22:43:26 GENSCAN_predicted_peptide_2 1003_aa MAPKKSTVLNVEQFIHDIERPAINRNFHCNKAFLQMWDELGAHKLPKIVL KAKWKGLRDNFRVEYKRIPRADNGDFMVDPATFESKWLHYYALLFLTDHMRHR LPKNEQDQSFYFSQSEDCEKTVVEPDLTNGLIRRLQDSEDEYDEEEMEADGEAS 30 EATMEETMPTPPAAHQMNQVSTTPLATGALRAQEEAHQHALIKAGLLRAQLMEL EKEAEDLSRKPPPPQMTSPVAPSLQVLVEPPAAHCSPPPMVTTTSAQVQPGSA AVLAPATTSASSVSSNGAPMGGKRSVSPPPLYNKAHPLATLAAHLAAKDRN EDFGPTSAVGGNGDHLSTQHSYANGLIPALKLKRPRLEDSNFNGSSTMDTLPVP EDDDYHYLLSLHPYMKQLTAAQKLRTKIQLFKELYKEDLEESNLDREVYVL 35 DDGAEVDLDLGNRYERFLDVTLHRDNNITGKIYKLVIEKERTGEYLGKTVQVVP ITDAIQEWVERVAQTPVQGSSKPQVCIVELGGTIGDIEGMPFVEAFRQFQFRVKRE NFCLAHVSLVPLPKATGEPKTKPTQSSVRELRGCGLSPLIVCRSEKPIGLEVKEKI SNFCHVGPDQVICIHDLSIYHVPLLEMEQNGVIEYLNERLQLNIDMSKRTKCLQQ WRDLARRTETVRREVCLAVVGKYTKFTDSYASVVKALQHAALAVNRKLELVFIE 40 SCLEETLHSEPSKYHKEWQKLCDSHGILVPGGFGSRGMEGKIRACQWARENQ KPLLIGICLGLQAAVIEFARNKLGKLDANTTEIDPNTANALVIDMPEHHTGQLGGT MRLGKRITVFS DGPSVIRQLYGNPKSVQERHRHRYEVNPKYVHLLQGMRFVG TDVDKTRMEIIELSGHPYFVATQYHPEYLSRPLKPSPPFLGLILASVDRLNQYIQRG CRLSPRQLSDASSDEEDSVVGLAGATKSLSSLKIPITPTNGISKSCNGSISTSDSEGA 45 CCGVDPTNGHK

>22:43:26|GENSCAN_predicted_CDS_2|3012_bp

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 gcgttgatcctaccaatggccataagtaa

40 **Human Homologue** TBLASTN with ORF1: CTP synthase (CTPS) (NM_001905.1)
Drosophila EST LD27370 (AA941993)

Annotated Drosophila genome genomic segment AE003532
Annotated Drosophila genome Complete gene candidate CG6854 - novel protein,
 possible CTP synthase?

45 **Human homologue of Complete gene candidate** gi4503133
 C33BD849A0044697
 [refNP_001896.1] CTP

synthase; cytidine 5-prime
triphosphate synthetase
[Homo sapiens] (8.40E-217)

5

Putative function

Enzyme important in the biosynthesis of phospholipids and nucleic acids, and plays a key role in cell growth, development, and tumorigenesis. The region of the human gene is the location of breakpoints involved in several tumor types

10

Confirmation by RNAi

Loss of G1 and G2/M peaks indicating fewer cycling cells

Example 35 (Category 4)

- 5 **Line ID** 225/27
Category Meiotic defects in testis: segregation defects
Reversion NR
Map Position 90D
- 10 **Rescue ID** 2D2P
Rescue Sequence 1
- Rescue ID** 2D2E
- 15 **Rescue Sequence 2**
GCCTGAACTTAAAACGCTGCCTTCGGCTCTCGCTCGGCACTCGCTCGGCTGCG
ACGTCGACTGCGACGCTGGCAGCGACAACAACGATTGGCCTCTCTCATTCACT
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25 CTACATTTTGAATGTTTCAATGAAAATTGCTGGGGAAGCTAGTGAACAACCA
TTTCGCCATAATTTACACTATCTAAGCTTTTATTTTAGCCACATGATATATGC
ATGCA
- 30 **Genomic hit, Accession No.** AC008361
- Associated ORF**
Genscan ORF1 predicted sequences >20:36:39|GENSCAN_predicted_peptide_2|515_aa
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35 DKFSKFAKLFHLQSKASVHLRETLVEALHYFTAPKVLVSDNERGLLCPTVLNLYR
SLDIDLYYAPTQKSEVNGQVERFHSTFLEIYRCLKDELPTFKPVELVHLAVDRYNT
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RDEPKSYGPGDEVFVANKQIKTKEKARFRCEKVQEDNKKNRNGKAAGGKGKTR
RVARGAQIYQNWAICRNLFSLACCRVCKVCDIVVEFRKGTNAVNVNQIREAI
40 SHVFHKEDIVIDVQESKEWCITWDDQVQSPLPELENLWHELWIGPSHAYLIDQIVD
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 caatttccctcatcgctctcgtagtcgtgatcatttcgggtatcatcatcattatcatcatgatcagcggttagcgcgccca
 15 aagaacaaatgccttttcacaccaccgatctcagcgatcagcgatcgcgcgacccctcaacaaaaagattcagcggtgcaaca
 ggtgcaggcacgatcttcggatgccttttgcagataccaccgatctccagggtccaggcgagccaacttattccgaagc
 caaatcgagaaattcttcgaaacgcgagtgccacaaaaattattgtttcgaattcgagccagtga

20 **Drosophila Gene Hit** BLASTN with rescue sequence: couch potato (Z14974).
Human Homologue BLASTX with couch potato: RBP-MS/type 2 (RNA binding motif
 family)(D84108)

25 **Annotated Drosophila genome genomic segment** AE003720
Annotated Drosophila genome Complete gene candidate CG18434 -couch potato RNA
 binding protein
 30 **Human homologue of Complete gene candidate** 2224621 dbj|BAA20798|
 (AB002338) KIAA0340
 [Homo sapiens] (2e-19) and
 Ensembl predicted peptide
 Gene:ENSG00000070877
 Clone:AC009710
 35 Contig:AC009710.00004
 (predicted unknown protein)

Putative function Possible RNA binding protein

Example 36 (Category 4)

Line ID 238/37
Category Meiotic defects in testis: segregation defects, multi-stage defects
5 (PI-02/17)
Reversion ?
Map Position 70D

Rescue ID I7E
10 **Rescue Sequence**
GTTCAAACGCACTTTTAAGGTGGCCTATCGGCCCATCAGGAGCAACTTTGTTC
TGCTGCCGGATCAGTACTACGGCGTCGTTTCGACCTATGTAAGTGTCTAAAGG
TCTTCGCTCCATTAGATTAGATACGCCAAAGATTAATCCGGTCAACCATCT
GATTAGGACACGGGCTGCCTGAGCTTGAGTACAATGGTCGGACGCACTACG
15 CCTCCTGGGCGCCACAAAAGGGCGGCGGCGGCATTAAAGACACCGAGATTGG
GATCAATGCCAGAGCGGCCAAGGAGATCGGTAAGCCATTACTTAACGGCCGG
ATGTGCATCGGTTGCCAATGTGCCGTAATATTGGACTCCGGCCATCTGCCCCG
TACCTCGTACGCTAGCAGCACCCACTTACCCTTTCTTGCCGTAGGTCTGCACGA
GAATGATCTGGTCAAGTGTGCGCTCATCGCTGACGTTCTAACCTGCGCAGCG
20 TCCACGTTACCCCCGTCTCGTCCAAGGACTGGGAGATCATAGTGAGTGACGGT
TTCGCTGCTTGCGGGCGTGG

Genomic hit, Accession No. CSC:AC017664

25 **Associated ORF**
Genscan ORF1 predicted sequences >15:26:30|GENSCAN_predicted_peptide_1|1819_aa
EMVQAKDPPSHYLSKLRITYLDPKASRSHRLYLFYFLCQKRKMVGESTSTQVLRD
LEISLRTNHIWVKEFLDDTNQGLDALVDYLSFRLQMMRHEQRLQGVLCASEERL
NLTNGGDGGEIVMGNSSSVSPGGGGLLSHGNSTGHGLANGTLDNRQHTMSYG
30 FLRPTIADALDSPSLKRRSRHIAKLNMGAAATDDIHVSIMCLRAIMNNKYGFNMVIQ
HREAINCIALSLIHKSLRTKALVLELLAAICLVKGGHEILGSFDNFKDVCQEKRRF
QTLMEYFMNFEAFNIDFMVACMQFMNIVVHVEDMNYRVHLQYEFTALGLDKY
LERIRLTESEELKVQISAYLDNVFDVAALMEDSETKTSALERVQEELEDQLEREIDR
NSEFLYKYAELESESLTLKTEREQLAMRQKLEELTVMQRMQLQHNEQELKKRDT
35 LLHTKNMELQTLRSRSLPRSASSGDGSLANGGLMAGSTSGAASLTLP PPPPPMPASP
TASSAAPPPPPPPAPPAPPPPPGFSPLGSPSGSLASTAPSPPHAPPM LSSFP PPPPPVA
GFMPAPDGA MTIKRKVPTKYKLPTLNWIALKPNQVRGTIFNELDDEKIFKQIDFNE
FEERFKIGIGGALRNGSNGTEVDGSLQSSKRFRPDNVSLLEHTRLRNIAISRRLG
MPIDDVIAAIHSLDLKKLSLENVELLQKMVPTDAEVKSYKEYIIRKDDQQLL TEED
40 KFMLQLSRVERISSKLAIMNYMGNFVDSVHLISPQVQSIAGASTSLKQSRKFKA VL
EIVLAFGNYLNSNKRGPAYGFKLQSLDTLIDTKSTDKRSSLLHYTVATIRAKFP ELL
NFESELYGTDKAASVALENVVADVQELEKGM DLVRKEAELRVKGAQTHLRDFL
NNSSEDKLKKIKSDLRHAQEAFKECVEYFGDSSRNADAAFFALIVRFTRAFKQH D
QENEQRLRLEKAAALAASKKENDQVLMRNKVNQKKQQEAVINELKSKAHSVRE
45 KKLLQQDEVYNGALEDILLGLKSEPYRRADAVRRSQRRRIDNNRLSRTLEEMDCL
HENDLVKCALIADV LNLRSVHVTPVSSKDWEIHELSTEKISGSVLEQTRIVNSTQILI

VWINKSMQVALTVDRCLKPHMNYGRIDHNTELVVAPNLKGLTNGTSNGVIEENT
 KLSRSKTTA QVKDELTEKL TPLTHSSTVSNVKNTIQNRKQDHMERLKKDLRRES
 SRSFEFRVIRGLWREQAQESDVFNKGHLPEFFDLDFYCMHTAADKDYVVRVR
 TVEDDIEDDLPETIHPSELNANLMKLLGIKELERVVLRPKTTVNVFVEKIELFANK
 5 KTHYKIMENAFKRFVIERTQHKPMLFNQEEVVRLDDLLVTVGILPEHFRYCVVD
 AQFLKESKIYAADLVRPVGEIIEETPPTSPLSVQDLIQLPEYDKIVDQVVQELRMN
 LCLSADNSVMRQCNVLLAGASGTGKTVLVERILDQLSRKPDYCHFEFFHGSRSKG
 RKTESIQLKDLRNIFTSCLQHAPAVVLENLDVLAHAAGEQSSQDGEYYNRMADTV
 YQLIVQYTTNNAIAVIATVNELQTLNKRLLSSPRGRHVFTVARLPNLERADREMLR
 10 ELCSHINVAKDLDLVKFSNLTEGYRKCDLVQFVERAIFYAYRISKTOPLLTNDQLI
 ESLEHTNSYCLQIQSNQRTGNDADANEMRVEELPGLESVVGVL EELVLMWPSRY
 PTIFNASPLRNQAGVLLYGPPGTGKTYLVSQLATSWNLRIISVKPELLAKYIGQSE
 ENVRNLFNRARSARPCVLFFDEFDSLAPKRGHDS TGVTDRV

15 >15:26:30|GENSCAN_predicted_CDS_1|5457_bp|
 gaaatggtgcaggcaaaggatcgccctcacattactgagtaaactgcgcacatatctgacccaaaggcatcaaggagtcac
 ggcttattctcttctacttcttctgtcagaaacggaaaatggcggcgagtcacgcccaggtgctccgcatctggagatctc
 gctgcgcacgaaccacatcgagtggtgaaggagttcctggatgacacgaaccagggtctggacgccctgctgactatctcag
 ctccgactgcagatgatgcgacacgagcagcgccctcagggtgtctgtgtgctcggaggagcgtctaatctcacaacggc
 20 ggcatggcgggtgagatagtgatgggaacagtagttctgttagtctggtggaggtggtggttactatcacatggaacagtac
 gggacatggtctggccaatggcacactgtactgaggcagcagcacacaatgtcctatgattcctacgacctaccattgccgat
 gctctgagatgctctagttgaagcgaaggtcacgacatattgcaaaataaacatgggtgccgccacggacacatccatgtgtc
 cattatgtgctgcgagctatcatgaacaataagtatgggtcaacatggttatccagcatcgcgaggccatcaactgcattgccttg
 agtcttatccacaatacgtcaggagcgaagccctggtcctggagctgctggcagccatctgtctggttaaggaggagacacgaa
 25 atcattttgggtcgttcgataatttaaggatgtgtgccaggagaagcgacgcttcaaacgctcatggagtactttgaactcga
 ggccctttaacatagattttatggttgcctgcatgcagttcatgaacatcgtgtccactcgggtggaggacatgaactacagggtgcac
 ttacagtacgagtttacagccctggccttgataagtatctggagcgaattcgattgacagaatcggaggaaactgaaggtgcagat
 atcagccctatttgacaacgctcttgatgtgtgctgcttgatggaggattccgagacaaaacttcagccctggaaacgagtcaca
 gcttgaggatcaactgagcgagaaatagatcgaactcagagttcctctataagtatcggaattagatccgagagttaacgct
 30 gaaaacggaaacgcgagcagctggctatgattcggcagaagctggaggaggaaacttacagtgatgcagcgaatggttcagcaca
 acgagcaggagctgaagaaaacgggacacactgctgcacacaaagaacatggagctgcagacgcttccgcttcctgcacga
 tccgctccagcggcgatggttctctggcgaatgtgtgctcctcatgctggttccacatcgggggagcctcttaacattgccacc
 acctccgccccaatgcccgcctcgcctactgcaagttcagctgctcctccaccacctccgccgagcaccaccggctccacc
 accaccgccgggctcagtcctgctggcagtcgcgagcggcagcctagcctcgacagcggccatcgccgccacatgccccgcc
 35 atgctaagctccttcaaccgccaccgcctcagtgccggctttatgccgctcccgatggcgccatgacctcaaacgcaagg
 tggccactaaatacaagttgccacctgaactggatagcactaaagcctaatacaggtacgtgtacaataattcaacgagctggatg
 acgaaaagatcttcaagcaaatcgacttcaatgagttgaggagcgcttcaagatcgggattggcgggtcttgcgcaatggtagc
 aatggaaacgaggtcgatgggtcgtgcagtcagcaaacgcttcaagaggccgacaatgtctcgtcgtgagcacacgag
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 cgcaaggaccaacagctactaccgaagaagacaagttatgtcagttgtcgtgtggagcgtatctgtcgaagctagcca
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 cttaagctgcaatcgctggacacgctgatcgatacaaatccacagacaagcgatcgtcactgttcactatattgtggccaccat
 45 acgggccaatttcggagctgctgaactcggagcggagctgtatggaacagacaaggctgcacggtggcactagagaatgt
 ggtggccgatgttcaggagcttgaaggggcatggtgtgcaaggaggccgagctgcgagtgaagggtgccagacg
 catatctgctgacttctgaacaacagcgaggacaagctgaagaagatcaagagcgatctcggcatgcacaggaagcggtc
 aaggagtgcgttgagtactttggcactcctcgggaatgcagatgcggctgcttcttgcgttgatcgtcgttcacgagagcg

ttaagcaacacgatcaggagaacgagcagcgtcttcgcttgaaaaggccgctgcgctggccgttccaagaagagaaacga
 tcaggtgcttatgcgcaacaaggttaaccagaagaagcaacaggaagctgtcataaacgagctgaagagcaaggcgcactcgg
 tgcgcgagaaaaagctgctgcagcaggacgaggtgtacaacggagccctggaggacatcctgctcggcctgaagagcgagcc
 gtacagggcgggcgatgctgtgcggcggtcgcagcgcggaggtatcacaataatcgtttatcgcgcacccctggaggaaatgg
 5 attgtctgcacgagaatgatctgtcaagtgtgcgctcatcgtcaggttctcaacctgcgcagcgtccacgttaccctcgtcgt
 ccaaggactgggagatcatagaacttagcactgaaaagatatcgggcagtgctggaacaaactcgcataatgaattcaacgca
 gatccttattgttgattaataagtcgatcaagttcgcgtgacagtggtatcgaagccgcacatgaactacgggagaataga
 tcacaatacggaaactcgtgtggcgcccaatctgtacaagggtctgaccaatggaactcaaatggtgttatagaggaaaacacaa
 aactctccagaagtaaaaccactgccaggtcaaggatgagctgactgaaaagttaacaccgttgaccattcctccacggtgtcc
 10 aatgtgaaaaatactattcagcgtacaagcgtcaggatcacatggagcgtcttaaaaggacttgcgcgcgaaagctcgcgta
 gcttcgaatttcgtgtcattcagggtctatggcgggagcaggccaggagtcggtatgtttgtgaacggaagcatctgcctgag
 ttctttgatctagatctattctatgcatgcacaccgcagccgacaaggattactatgtgagagtgcgcacagtagaagacgatattg
 aggacgatctaccagaaaccattcatcatcgatgaactaatgccaatctatgaagttgctgggtatgaagaaatggaacgag
 tggttctaagacctaaaactaccgtagttaactttgtagaaaaaattgagctatttccaacaagaagacgcactacaaaatcatgga
 15 gaacgcatttaagcgatttggatagagagaactcagcacaaagccgatgctcttaaccaggaggaggtgtacggcgtggagga
 cgattactggttactgttggaattttaccagaacacttctgttattgcgtggtggagcgcagtttctgaaggagtcgaagatctacg
 cagcagatctggtgcgtccggttggcgagattattaaggaggagacgctccgacatcgccactaagtttcaggatctcatcca
 gttaccggagtagataagattgtggatcaggtagttcagggaattgcgaatgaatctatgcctcagtgctgacaattccgtatcgt
 20 cagtgcaatgtctactcgtggtgcctcgggaacgggtaaaacagttctgtggagcgcattttggaccagctgtcacgcaagcc
 ggattattgtcacttcgagttcttcacggatcgcgaagcaaggccgcaagacggagtcctccaaaaagatcttcgcaacatttt
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 ccaccgtcaacgaggttgcagaccctcaataagcgattgagctcaccaagggaagacatgtctccagactgttgcgtctgccc
 25 aatttggaaacgagagatcgagagataattcttcgagagctgtgcagccatatcaatgtggccaaggacgtggttgaagtct
 ccaacctcacggagggtaccggaaatgtgatctgttcagttcgtggagcgtgcaatattttatgcttatgcataagcaagacc
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 caggtaaaacctatctggtctctcagttggccacatcgtggaacctgcgcacatttccgtcaagggtcctgagttgctgccaaata
 30 tattggtcaaaagcgaggaaaatgttcgaaacctgtcaatcgagctgcagtgcccgaccatgtgtgctttctcagcaggttgac
 agcttggcgccgaaacgtggtcacgattccacgggggtcaccgatcgagtg

Drosophila Gene Hit recue sequence and TBLastn with ORF1: mRNA for l(3)70Da (AJ243811)

35 **Human Homologue** BLASTX with l(3)70Da: peroxisome biogenesis factor 1 (AF026086)

Drosophila EST LD43687 (AI512050)

40 **Annotated Drosophila genome genomic segment** AE003536

Annotated Drosophila genome Complete gene candidate CG6760 mRNA for l(3)70Da
 - novel protein with
 homology to endoplasmic
 45 reticulum ATPases

Human homologue of Complete gene candidate 4505725

ref|NP_000457.1|pPEX1|
peroxisome biogenesis factor
1 >gi|2655141 (AF026086)
(8e-80)

5

- 10 **Putative function** Putative member of the AAA protein family (ATPases associated with diverse cellular activities) including homologies to transitional endoplasmic reticulum atpases, and an E.coli membrane-bound AAA-type metalloprotease which degrades degrades sigma32, an alternative sigma factor for heat shock promoters
- 15 **Confirmation by RNAi** Slight loss of G1, increase in G2/M indicating arrest in G2/M

Line ID 238/44
Category Meiotic defects in testis: segregation defects, multi-stage defects (PI-02/18)
Reversion R
Map Position 70D
Rescue ID F8E
Rescue Sequence
 GTTCAAACGCACTTTTAAGGTGGCCTATCGGCCCCATCAGGAGCAACTTTGTTC
 10 TGCTGCCGGATCAGTACTACGGCGTCGTTTCGACCTATGTAAGTGTCTAAAGG
 TCTTCGCTCCATTAGATTAGATACGCCAAAGATTAATCCGGTCAACCATCT
 GATTAGGACACGGGCTGCCTGAGCTTGACGTACAATGGTCGGACGCACTACG
 CCTCCTGGGCGCCACAAAAGGGCGGCGGCGGCATTAAAGACACCGAGATTGG
 GATCAATGCCAGAGCGGCCAAGGAGATCGGTAAGCCATTACTTAACGGCCGG
 15 ATGTGCATCGGTTGCCAATGTGCCGTAATATTGGACTCCGGCCATTTGCCCCG
 TACCTCGTACGCTAGCAGCACCCACTTACCCTTTCTTGCCGTATGTCTGCACGA
 GAATGATCTGGTCAAAGTGTGCGCT

20 Other results same as for line 238/37

Line ID 428/5
Category Meiotic defects in testis: cytokinesis defects, segregation defects (seg-01/01)
Reversion ?
Map Position 70A
Rescue ID G4E
Rescue Sequence
 30 GTTCAAACGCACTTTTAAGGTGGCCTATCGGCCCCATCAGGGAGCAACTTTGTT
 CTGCTGCCGGATCAGTACTACGGCGTCGTTTCGACCTATGTAAGTGTCTAAAG
 GTCTTCGCTCCATTAGATTAGATACGCCAAAGATTAATCCGGTCAACCATTC
 TGATTAGGACACGGGCTGCCTGAGCTTGACGTACAATGGTCGGACGCACTACG
 CCTCCTGGGCGCCACAAAAGGGCGGCGGCGGCATTAAAGACACCGAGATTGG
 35 GATCAATGCCAGAGCGGCCAAGGAGATCGGTAAGCCATTACTTAACGGCCGG
 ATGTGCATCGGTTGCCAATGTGCCGTAATATTGGACTCCGGCCATCTGCCCCG
 TACCTCGTACCTAGCAGCACCCACTTACCCTTTCTTGCCGTAGGTCTGCACGAA
 AATGATCTGGTCAAAGTGTGCGCTCATCGCTGACATTCTCAACCTGCGCA

40 Other results same as for line 238/37

Line ID 848/7
Category Mitotic defects in brain: cytokinesis defect. Meiotic defects in testis: cytokinesis defect. Multi-stage defects
Polyploidy, no overcondensation
5 **Reversion** R
Map Position 70D1-2

Rescue ID G1E

10 **Rescue Sequence 1**

GGCCACCTTAAAAGTGCGTTTGAACATTCTCGTCGTGGGCGTGTGCGAATTTA
GTACGCTCCTTCCTGGTTTAAATCATTTTCGCACTAAACTTCTGCTCTCAGCGG
AATTTACTTTTGCTTTATTAGAGATGGGAGCTCGCGCATCAGCTGAGCCGATA
CTTGCGCAACAGGTGATACAGCTGATTAGAGATGGCCCTTTTCAACTGTTCCC
15 AGCAGTGACCGCTGCCATAACCGTTTTTCAAATTTACGTGAGAACAGACATAA
AATAAATATTACAGCTCGTAGTAAATGTTATTCTATATTTAAAAGGAAATTGT
AATAGTTAAAACTTGCAATGAATCAGTTACGTTCAAAAAAGGAAACACACTTT
AGTTTTTGGCTAGTTTATTGGGTAAATAATTTTTATTTAAAATAGTTCGAGTG
TTCAATATAGTCATGTAAATGTGTACAGAAAGATCCGGCATTTGATATTTAAT
20 ATATCGATTTCCCTTCACTTTCGCTCCTCGTATACCATGCTGGGGTCTTATCAA
ATTTATT

Rescue ID G1P

Rescue Sequence 2

25 AAGGTGGCCTATCGGCCCATCAGGAAGCAACTTTGTTCTGCTGCCGGATCAGT
ACTACGGCGTCGTTTCGACCTATGTAAGTGTCTAAAGGTCTTCGCTCCATTCAA
ATTAGATACGCCAAAGATTAATCCGGTCAACCATTCTGATTAGGACACGGGCT
GCCTGAGCTTGACAGTACAATGGTCGGACGCACTACGCCTCCTGGGCGCCACAA
AAGGGCGGCGGCGGCATTAAAGACACCGAGATTGGGATCAATGCCACAGCGG
30 CCAAGGAGATCGGTAAGCCATTACTTAACGGCCGGATGTGCATCGGTTGCCAA
TGTGCCGTAATATTGGACTCCGGCCATCTGCCCCGTACCTCGTACGCTAGCAG
CACCCACTTACCCTTTCTTGCCGTAGGTCTGCACGAAAAATGATCTGGTCAAG
TGTGCGCCTCATCGCTGACGTTCTCAACCTGCACAGCGTCCACGTTACCCCCGT
CTCGTCCAA

35

Other results same as for line 238/37

Example 37 (Category 4)

Line ID 252/40
Category Meiotic defects in testis: segregation defects, abnormal spindles.
5 (Ab-03/30)
Reversion R
Map Position 84E

Rescue ID A4B
10 **Rescue Sequence 1**
TACATGACTCTGCGATTTGACAAAAACAAAATTGAGTTTTGTCAAGAAAATCA
ACTATTTTTCTGTGTTTAAAAAACCGAAGCCAACAAATCCGACCAAAATGCCT
GCCGAAAACCTGGAGGAGCAGGGTCTGGAGAAGAACCCGAACCTGGAGCTGG
CCCAGACGAAGTTCCTGCTTACCCTGGCGGAATACAAGCAGGATGCGGCATTG
15 AAGGCGAAGCTTCTGGAGGCGATTGCGACGGAGAATATGGCCCCGTGGGTAC
GAGCACATCCTGCTCCGGAACCTCGGCTTGGACCCGTTAGACAAGGATCTTGCC
TGGCGCCGAATTGAAGGAAAAACAATCGCGTTTAAGTTGGGAGCCA

Rescue ID A4E
20 **Rescue Sequence 2**
GTCATGTACTACCAGTGTGACCCCAAAGTTATCGATAAATTATACCGCATATT
TTAACATTGCCAAAAATACCAGAGCGATGTCCATCAAGATAGCGACGAAATT
AGAACAGTGCAATTGCCAATTGGGAATTTGTATTTTAATTTATTTTTAAATTCT
GAAAGTAATTTTAATTTAAAAAAAACCTTGAGAGCTGTCTAGAAAAGAACTTAT
25 GTTTCATGATAACTTTGTCGAAGAATTAAGAAATATTTAGTTGTAAAATAATT
GTNTGAATCTATTTTTTTTCCAATAACACGACTTATATTTTTTTTTTAAAATATTC
CGAGCTAAATCCCAAGAAAGTTAACTCCAATCTTGGGATTTTGAAGTGCCCC
AGAACTCCAAATTAACACTTCCTTTTTTAAATAATTGTTAAGACCCGTATCA
CTTATGGTTATATACTGACCTCGAAAGGGCCACACTAAGGGGGGAGTTTGAAA
30 ATTGATTTTCTGATAAAAAATTTTCGTTGGAAGCTACAGCATCGTCCACTGTC
CATGTTTATATATCCTTATATTTCCTATAAATATAT

Genomic hit, Accession No. AC006494

35 **Associated ORF**
Genscan: ORF1 predicted sequences >23:00:28|GENSCAN_predicted_peptide_2|389_aa
MPAENLEEQGLEKNPNLELAQTKFLLTLAEYKQDAALKAKLLEAIRTENMAPWY
EHICSELGWTVDKDLLARMKENNRVEVEQLDAAIEDAEKNLGEMEVREANLKKS
EYLCRIGDKAAAETAFRKTYEKTVSLGHRLDIVFHLIRLGLFYLDHDLITRNIDKA
40 KYLIEEGGDWDRRNRLKVYQGVYSVAVRDFKAAATFFLDTVSTFTSYELMDYPT
FVRYTVYVAMIALPRNELRDKVIKGEIQEVLHGLPDVKQFLFSLYNCQYENFYV
HLAGVEKQLRLDYLIHPHYRYVREMRLGYTQLLESYRSLTLQYMAESFGVTVE
YIDQELARFIAAGRLHAKVDRVGGIVETNRPDNKNWQYQATIKQGDLLNRIQKL
SRVINI

45 >23:00:28|GENSCAN_predicted_CDS_2|1170_bp

atgcctgccgaaaacttgaggagcagggcttgagaagaaccgaacctggagctggccagacgaagttcctgcttaccc
 ggccgaatacaagcaggatcgggcattgaaggcgaagcttctggaggcgattcgacggagaatatggccccgtggtacgag
 cacatctgctcggaactcggctggaccgtagacaaggatctgctggcgcgaatgaaggagaacaaccgcgtagagggtggagc
 agctagatcgcgcaatcgaggatcgggagagaagaatctggcgagatggaagtgcgcgaggcgaatcttaagaagtcagagta
 5 cttgtgcccatcgcgacaaggctgccgagagactgcctccgcaagacctacgagaagaccgtttccctgggtcaccgcct
 ggacatcgtgttcaatcgatccgcttgggactgtttaccttgaccacgatctcatcactcgcaacatcgacaaggccaagtatctg
 atcgaggaaggcgcgattgggaccgacgcaaccggtgaaggctaccagggtgttactcgggtggcggtgcgtgacttcaag
 gcggcgccacgttctcctggacaccgtaagcaccttcacctacacgaactgatggactacccaccttcgtgcgttacaccgtt
 tacgtggccatgattgccctgccgcgaatgagctgcgcgacaaagtgatcaagggtccgaaatccaggaggtgtccatggc
 10 ctgcccgcgtgaaacagttcctgtttcctgtacaactgccaatatgagaacttctacgtacacctggccggcgtagagaagcaa
 ttgcgttgactacctcattcatcccaactaccgctactacgtgcgcgagatgcgcattctgggtacaccagttgctggagtcg
 tatcgtccctcaccctgcagtatatggcggagtcgttcggcgtaacagtgaatacattgaccaggagctggcacgcttcacgc
 cgccggacggctgcatgccaagggtgacgcgttggcgcgattgtggagaccaatcggcctgacaacaagaactggcagttacc
 aggcgaccatcaagcaggcgatctgctgctcaaccgcatccagaagttgagccgctgataaacatctaa

15

Drosophila Gene Hit BLASTN with rescue sequence 1 and TBLASTN with ORF1: 26S proteasome regulatory complex subunit p42A (AF145308).

Human Homologue BLASTX with EST and TBLASTN with ORF1: Hypothetical protein KIAA0107 (D14663).

20 **Drosophila EST** several including GH17651 (AI387197)

Annotated Drosophila genome genomic segment AE003739

25 **Annotated Drosophila genome Complete gene candidate** CG5378 - Rpn7 19S proteasome regulatory particle, non-ATPase protein, subunit S10aHuman Homologue

30 **Human homologue of Complete gene candidate** gi7661914
8843E6684AE91ACD
[refNP_055629.1] KIAA0107
gene product [Homo sapiens]
(3,40E-149)

35

Putative function component of the 19S proteasome regulatory particle

Confirmation by RNAi Marked decrease in G1 and G2/M indicating fewer cycling cells

40

Example 38 (Category 4)

	Line ID	277/7
	Category	Mitotic defects in brain: anaphase defects (weak, higher condensation, some polyploidy, fewer anaphases, polyploids with monopolar spindles)
5	Reversion	?
	Map Position	71B
	Rescue ID	B8E
10	Rescue Sequence	AGTCGGCGCATGCGGAGAGAGAATCGAAAGAGAAAGAGAAGCAAAGAGAGC GACATACAGCAAAAACAATTCAAAAAGAACTGGTGAAGAATACGAAAATAAG ATAATTTTTTAAAGGAAGTCGCGCTTTGATCCGTATCCGTTTTAGCGTCCAAGAT TTATATCTTAAATCGGACCTATATTTTGAGGTACAGTGAAGCTTTGATGCGCCA 15 GTCTTATATGAGTTAAAGTTTTAACGATTGAAAGACACCCCTGAGCTGCTCAT TATATTTCAATATTTATAAACAATCTTATATCAGAGCTTGAGAGACTTGCAATGC GCCACAAAATTCCAATTCCAATTCCAATCCGGAATAATTTACAATAATCTC AATTAACATACGTATTTTATGTTTCGTAATTTTTTAAAATTCCCAGATTCCCCAC AATTGCCATAATAATCTCGATTATGTTATTATACTCTGAGAAGTAGGAGTGTG 20 TGCAAAGACCACAAACAAATCATTAGGGGCGT
	Annotated <i>Drosophila</i> genome genomic segment	AE003584
	Annotated <i>Drosophila</i> genome Complete gene candidate	CG15383 – novel
25	Human homologue of Complete gene candidate	none
	Putative function	No homologies to indicate function
30	Confirmation by RNAi	Slightly increased G1 decreased G2/M indicating arrst in G1

Example 39 (Category 4)

	Line ID	284/4
5	Category	Mitotic defects in brain: anaphase defects (overcondensation, polyploidy (with overcondensation), few anaphases, metaphase with bipolar spindle)
	Melotic	
	Reversion	NR
10	Map Position	89B
	Rescue ID	2C6E
	Rescue Sequence	GTCTACCACTAGCTCTTTGTCTTCGCCTTCTAGTCTCTCTCATCTTGGCAGCCC GTTCTAGTGCGCGTATTTTTAGTCGCAACACATTGCCCAATTCGCCAGCCGCTA 15 TTTGTGTCGTCCATTTGTTCAATTCATCGGGCTCTTTTCCGATTTCACTGGGTGG CATTAAACAATAATCCCTGCGTTCGCTGTCCACGTCCACATTACGATACGTTTA GTGCACGGAAAGAAATAAGCGTGTGGTTTCATAATATTAGCTATTGAAAAAA GTTCTTAAATTTAAGCCTCACTCGATTCTGATGCATGAAATATTATTGGATTGT AAATGAGCGTCATGTTTTGGTATACAAATCTCAAAGTAATTTAAAAATTCTCA 20 TCTTACCGTACCTTGAACCACTACCAATCATCTCAGTACAGCATTTACGCGAA TTTCTCACTGTGCACTACAATGCCAGGCGGTACAAGCACCTGTATTTATTATG GTCCGCTGCCGTAATCGACTGCAGTCGCCGCTTCCCTCTCTTTTGCTACCAA CAACTTGGGGTAGGGCACCTGAACTAGTTTCAAACGGCGGCGGTCCGCCTTTT CAGCTTTTTCGCATTTGCCATTTTCCCGCGG
25	Annotated <i>Drosophila</i> genome genomic segment	AE003711
	Annotated <i>Drosophila</i> genome Complete gene candidate	CG4275 - mor transcription factor involved in chromatin remodelling
30	Human homologue of Complete gene candidate	CG4275- 4507081 [ref]NP_003066.1 pSMARCC 2 SWI/SNF related, matrix associated, actin dependent regulator of chromatin, subfamily c, member 2(aa)
35		
40	Putative function	Transcription factor, regulator of chromatin
	Confirmation by RNAi	Decrease in G1 and G2/M and increase in polyploidy

Example 40 (Category 4)

Line ID 407/8
Category Meiotic defects in testis: cytokinesis defects
Reversion ?
Map Position 64B1-2

Rescue ID A9E
Rescue Sequence
 10 GACTCACCCTTTCACGCATTTTCATTGGAACGTTTGTTCGTTTATGCACACGC
 GTGTTGACACTTTTCATGAAACGCAGTGCCTGAAAAGTGCATCGCATAAACGC
 AATAAATGTTTGATGGATGCGTTCTGATGGCTTGAAGTCGCCTATTTGGCCGA
 TTTTCGCACGTCCACTCCCGACGGCAACAGAGTCCTGACTGAATCCCGGAGCG
 GAAGGAGTGTGGATAGCCAGGACTGCCAAAGGACACTGCGCACTTTTACTTTT
 15 TCGAAAGCGAAAGCGAAAGTGGTGGGGCCCAGGCCAAAACAANCCCTTGAGT
 TGAAATTGGAAAAAAACCGGGACAGGGATGGGAGCCCAGCTCCAACAAACG
 GTTCCGGATTCTTGGGAAAGCCACGCCCTGCGCCTGGAAAAGGAATGCCCTC
 CACCTCATTTGTCTCCGTTTTGCGCTATCTCTCCCCCAAATTTCCGTTAAATG
 AAAAACAACTTTGGGTTTTTGGTTTTTAACAATTTCTCCCCATTTGGTTTTNGGG
 20 TTTCCCTTTCCATTTTTTGAATTGGTTTTAATTAAAT

Genomic hit, Accession No. AC005814 64A6-64B6

Associated ORF
 25 Genscan ORF1 predicted sequences >22:57:22|GENSCAN_predicted_peptide_2|524_aa
 MGRRKDKPRVPEQDARICRAICLCQLTMVLSCVSIVYLSVAIYSPSLKAFKSGFEL
 DPVMCQTVDRQMPNCPWASCGEWCLTKTSGFCPQIHSIVRRNGTDIQLNNCTR
 VTNTSCAMIDLSRLNKFNCNNGTACNNIRGVFNCSNGHCKNMSEFFLCHHKADG
 LTVNSQKDNTKLNGLFFECHGVHCTKIKKPFSCDRYCSKITTTNVNTLIMHEDNLIA
 30 ADCENAVAFNQARGSEHGVRIEPFEFWKEDDGNLLTNCATVTRESNDRITATDCI
 NGTLLEHDTLPAPFMNFTQFWAIYENSTRSDPEQRYLPNQANLTIYSWKKLFINL
 EGCVNTLRGECKDFVARYGNDGDNNTAQSR YQCYYNKDSNVEFVVAR YDLDK
 VYRELLVSLVPIVLFVISSISLCITKS VKVGDDAKMRCVCAGDSDNDGPFPGPL
 ANKQQDQMYD TDDDVVDLEHQA VDGQELSDHGLPLDNQELIGSTKSLIPSPVGE
 35 SGTSDQIFDQDQEKATTCDVPEKPLVIL

>22:57:22|GENSCAN_predicted_CDS_2|1575_bp
 atggggcggcgcaaggacaaaccggtgattccgaacaggatgcgcacatctgctgcgccatctgcctgtgccagctgac
 catggtgctgtcctgctgctacacgaagcgtggccatctactgccctccctaaaggccttcaagtcggcttcgagct
 40 ggatcccgtcatgtgccagacggtggtgacgacagatgcccaacaactgccctgggcatcctgcggcgagtggtgctgacca
 agaccagtggtctttgccccagatccactcaatagtcgctgcaacggcaccgatccagctgaacaactgcaccagagtcac
 caacacatgctgcgccatgattgacctgagtcggctgaacaagttcaattgaacaacggcaccgcctgcaacaatatcaggagc
 gtcttaactgctccaatggacactgaagaatatgctggagtctctgtgtcaccacaaagccgatggacttacggtcaattcgc
 agaaggataacaccaagctgaatggattcttcgagtgacggggtgactgcaccaagatcaagaagcccttcagctgcgatc
 45 ctactgttccaagataacaactaccaatgtgaacaccttattatgcacgaggataatcttattgccgccgattgtgagaacgcagtg
 gcttcaaccaagcccaggatccgagcacgggtgtgcgtatcgaacctttagtggaaagaggatgatggcaacctgctga

ccaactgcgccacagtcaagagagtcggacaatcgcatcactgccacggactgcataaatggaaccctcctggaacatgaca
 ccttgcccgtcccttcataacttcacccagtttgggccatctatgagaacagcaccagggtcggtggatcccagcagaggtac
 ctgccaaccaggccaacctgacctctacagctggaagaactgttcataacctggagggtcgtgtaacacactgcgtggg
 gagtgcaggactttgtggctcgtatggcaacgatggcgataacaacaccgccagtcacgctaccagtgtactataacaagg
 5 actcgaatgtggagtttgtggtgcacgctacgattggacaagggttacagggagcttctagtctcgtgattgtgccattgtgctc
 ttgtgatctcatatatcgttatgtatcatcaccaaatccgtcaagggtgggtgacgatgccaagatgcgctgtttgtccggcga
 tgattcagataatgatggccctttggccaggactagcaacaagcagcaggatcagatgtacgatacagacgacgatgtagt
 gacctggagcaccaagcgggtggatggtaagaactatcgaccacggacttccgctggacaaccaagagctaatacggtagcac
 caagtcgttgataccaatcagtcctcggaagaatccggaactagtgtatcaaatcttgaccaggatcaggagaaagcaactacgt
 10 gcgatgtcccagaaaaccactagtcatactataa

(corresponds to CG15003)

Annotated *Drosophila* genome genomic segment AE003480
 15 Annotated *Drosophila* genome Complete gene candidate CG15003- novel unknown

Human homologue of Complete gene candidate none

20 Putative function No homologies to suggest function

Confirmation by RNAi Only wild type profiles observed

Example 41 (Category 4)

Line ID 422/28
Category Meiotic defects in testis: segregation defects, multipolar spindles
 5 **Reversion** NR
Map Position 68E

Rescue ID 2I4E
 10 **Rescue Sequence**
 TCGTGGACCCTCAAAGNAACGGATTTCTCCAGTTTCTTCAAAGGGTTAATAAA
 CTTTTCGCACGTTTCGCATTTTTATGCTCAATCCGGTTACAAAATGCTGATAAA
 ACCACTTGAACACACGTTTCCGTACTGATAAGGGCTTTTCTTCTTATCTGACC
 TCTGGAATCCGCGGAATTAATTCTTGAAGACGAAAGGGCCTCGTGATACGCC
 15 TATTTTATAGGTTAATGTCATGATAATAATGGTTTCTTAGACGTCAGGTGGCA
 CTTTTCGGGGAAATGTGCGCGGAACCCCTATTTGTTTATTTTCTAAATACATT
 CAAATATGTATCCGCTCATGAGACAATAACCCTGATAAATGCTTCAATAATAT
 TGAAAAAGGAAGAGTATGAGTATTCAACATTTCCGTGTCGCCCTTATCCCTTT
 TTTGCGGCATTTTGCCTTCCTGTTTTTGCTCACCCAGAAACGCTGGTGAAAGTA
 20 AAAGATGCTGAAGATCAGTTGGGTGCACGAGTGGGTACATCGAACTGGATCT
 CAACAGCGGTAAGATCCTTGAGAGTTTTCGCCCCGAA

Genomic hit, Accession No. CSC:AC014962

25 **Annotated *Drosophila* genome genomic segment** AE003543
Annotated *Drosophila* genome Complete gene candidate CG5684 (putative
 transcription factor, human
 homolog
 30 **Human homologue of Complete gene candidate** 1e-100 4758946
 ref|NP_004770.1|pPOP2|
 POP2 (yeast homolog)
 >gi|4106061|gb|AAD02685|
 35 (AF053318) CCR4-associated
 regulator of polymerase II
 transcription

40

Putative function Transcription factor

Example 42 (Category 4)

Line ID 422/5
Category Meiotic defects in testis: segregation defects, abnormal spindles
 5 (Ab-04/26)
Reversion ?
Map Position 82D

Rescue ID B9E
 10 **Rescue Sequence 1**
 ATTTGGCTCTTGATGGACTACAACGCTACCAAAATGGGGCTTGAGTTGAATTAC
 CTGTTGGAAGACACAATGCCACCCACGATCAACAATTCGGCGGTAAACAGTG
 CCGCCGAAAAGCGACCCAGCGGCAAACGGGAGCGCAAGTAAGTGAACAGAT
 CCCTAAACAGACCCAGATACTCAGACTGATGTGTACCTTGCAGATCCGAGATC
 15 ATTTGCCGCGTGAAGTATGGAAACAACCTGCCGGATATACCATTTGATCTGAA
 GTTTCTGCAGTACCCCTTCGACAGCCACCGCTTCGTGCAGTACAACCCAACGT
 CGCTAGAGCGTAACTTCAAGTATGACGTGCTGACGGAACACGATTTGGGTGTC
 ACGGTGGGACCTGATTAACCGGGAGCTCTATCAGGCCGACTCCATGACGCTGC
 TGGACCCGCCGATGAAAACTGCTGGAGGAGGAGACTCTGACGCCACAGAC
 20 TCTGTGCGTTCGCGCCAGCATTTCGAGGACGGTGTGATGGTTGCGCAAATCCGA
 GT

Rescue ID B9B
Rescue Sequence 2
 25 GGCCAAATCTAGAAATCCTCAAATCTGCGCTTGGCAGTGTGACCGTACTTGAC
 CGGTACGATAATACCTCCGGTAAAAAAATACTATATTTCCGGGGGACTCAAA
 TGCAACATCCTCATCGTATATAACACAACATCTATTTGAATTTTCAATTTCCACAA
 CTAATATTATGGATAATGCTTTATTATCATTTTCCAAGTTAGCGATAAATCACC
 CCACAAGCTGAAAAATCAACGTTTAAAAACGATTGATATTTTTTTAATACTTT
 30 TTGGTTTTACTATTTGAATTTTTGTATACTTTTAGATTTTACTATTTTAATTTTTT
 GTTTCTTCTAGCTGACTAACGGGTTAAAAAAGGATCCGTCGACCTGCAGATCT
 CTAGAAGCTTGCGTTGCTGGCGTTTTTTCCATAGGCTCCGCCCCCTGACGAGC
 ATCACAAAAATCGACGCTCAAGTCAGAGGTGGCGAAACCCGACAGGACTATA
 AAGATACCAGGCGTTTCCCCCTGGAAGCTCCCTCCGTGCGCTCTCCTGTTCCG
 35 ACCTGCCGCTTACCGGATACCTGTCCGCCTTCT

Genomic hit, Accession No. AC008189
Associated ORF
 Genscan ORF1 predicted sequences >15:53:24|GENSCAN_predicted_peptide_3|211_aa
 40 MRNANESSGPKPSKFVSNEFHAFSTICSIADSPAVSREKLKIDLAARKIPSASAPK
 GDSPLERFSRDLFTYLRVCRWGRFSAALFTAELLIVGGIVSSNRTSESSETGNPLA
 NEPDPLYMKLVDPMPVAGESPKRMIKDQKDVGLKSTSSSEELRKLKPKTRGRQKRFI
 RNPNYVKANEFYDKMLSSEYVSKRYKDLPPPHPGFGADQPPA

45 >15:53:24|GENSCAN_predicted_CDS_3|636_bp
 atgcgcaacgcaaatgaatcgagcggtaaaccaaaatcgaaattgtaagcaacgaattccacgcattgtttcaacaattgttcaa

146

ttgccgattccccggctgtctctcgagaaaaattgaaatcgatttagctgctcggaataccttcggcatcagccccaaaggg
gattctccactcgagcgcttttcgcggaatctgttcaacttactgcgctccgttggcgtggggtcgcttttcggcggcactgtttacc
gccgaattgtgatcgtgggtggcattgtgtctccaacagacgtcagagtcttctgaaactggaaaccacttgcaaagagccc
gatccattatatgaaactgggtgatcccatgtagcaggagaaatcacctaaaaggatgattaaggatcagaaagatgtaggcctt
5 aaatcaactagcagtagcgaagagctccgaaaattgccaaaaacgcgaggtcgacagaagagattcattcgggaatccaaactat
gtgaaagctaacgaattctatgataagatgttaagcagtgaatacgttaagtaagcgggtataaggatctccgccgcctcatccggga
ttggagcggatcaaccgccagcatga

Corresponds to CG2503

10

Annotated *Drosophila* genome genomic segment AE003605
Annotated *Drosophila* genome Complete gene candidate CG2503 - novel possibly
RNA binding

15

Human homologue of Complete gene candidate 3287674 AC005239
(AC005239)
F23149_1(aa)

Putative function Possible RNA binding protein

20

Confirmation by RNAi Almost no G1 and broadened G2/M indicating arrest in
G2/M

Example 43 (Category 4)

	Line ID	423/14
	Category	Meiotic defects in testis: cytokinesis defects, abnormal spindles
5		(Ab-16/13)
	Reversion	R
	Map Position	67B1-10
	Rescue ID	E9E
10	Rescue Sequence	
		GTTTGGCGTAAAAGCTTCGGCTGTGTTTGGTGCCCAAATTTTCCACTGCTTCT
		CTTTTGTGTATCTCTTATATCTTGTGCTTTTTTGTGTGTATGTTTTCTCGTTTC
		TTTTTGCACACGCGCTTCGCGTTGCGGGCCAGCTGTTTTGTTTGATAAGTGGT
		TACGGTTTGTGTGTGCCAGCGGGTTTTCTTAGTCGAACTGCTCGCGATGACTG
15		ATTTTTCACAAGTGACTCAAAAACAGTCGATCGCCCTTTTAAGAAAACCCGCT
		CAACGCACACAAAAGCGGTTTCTCTCTTTTTGTCTGTTCTCTCTTTTCACACTGA
		CCACACGGAACGAAAAAATGATTACCGACCACACGGAAAGAAAAATTTATGT
		CCAGACGAAACTATTTTTGTCCAAGTAGCTGATTTGCATAACAATTTAAGCCA
		CAAGAACTAGATTAAAAATTTTACATTTAAATACATTATCAAATCCGAAATAT
20		CAATAATTGTAATTTATCCTTACAAAATGTTA
	Genomic hit, Accession No.	CSC:AC020214
	Drosophila EST	several including LP12306 (AI297868)
25	Annotated Drosophila genome genomic segment	AE003552
	Annotated Drosophila genome Complete gene candidate	CG3967 - novel
	Human homologue of Complete gene candidate	none
30	Putative function	No homologies to indicate function
	Confirmation by RNAi	Only wild type profiles observed

Example 44 (Category 4)

Line ID	427/5
Category	Mitotic defects in brain: anaphase defects. Meiotic defects in testis: segregation defects, abnormal spindles (mitotic : Overcondensation, lagging chromosomes/less aligned metaphase with bipolar spindles, Meiotic: Ab-06/20)
5	?
Reversion	?
Map Position	67B1-5
10	Rescue ID H4E
Rescue Sequence	GTACAGCCTGAAGTGATCGTTGTTGTTTGAATCGGTGCTATCGGCGGTTGCGC TTTGTGGGCATCTTTATCCAATTTGCTATGCGCGCTTGTCCTTAAATTTTGAAC TGTATTCCAAGGGTTGCTTTGGCGGCTATCGATAGTATCGGCATGGTTACATT 15 TAGTTTTATAACAAGAATTTTACAGGTATTTGATTATCTGAGCTTAGTTTTAA GCAANAATATTATTGTTAAAAATTTAAAAAGTAAACAAGCTATTTTAAACAAGC ATTTAAACAAATAGTATTAATAATATAAAAATATATCGATATGTGTTGCAAAT GTTCGTTCCCTTAGTATTCTCTCATATTTATTTCAAATAAACTGTATAAAATAT CTGAAAAAGCGAACATATTTATTTAATTTTCATCGCAGATATCGATATCACAGC 20 GCTGCTATCGATGGTGTGTCTGTCGCAGTGCCTATCGCTTACCCTGCCATCGCT AACAAAAA

Genomic hit, Accession No. CSC:AC020120

25	Associated ORF
	Genscan: ORF2 predicted sequences >22:06:07 GENSCAN_predicted_peptide_7 464_aa MPSEQHTNIKVA VRVRPYNVRELEQKQRSIIKVMDRSALLFDPDEEDDEFFQGA KQPYRDITKRMNKKLTMEFDRVFDIDNSNQLFEECTAPLVDAVLNGYNCSVVF YGATGAGKTFTMLGSEAHPLTYLTMQDLFDKIQAQSDVRKFDVGVSYLEVYNE 30 HVMNLLTKSGPLKLREDNNGVVVSGLCLTPIYSAEELLRMLMLGNSHRTQHPTD ANAESSRSHAFQVHIRITERKTDTKRTVKLSMIDLAGSERAASKGIGVRFKEGAS INKSLALGNCINKLADGLKHIPYRDSNLTRILKDSLGGNCRTLMVANVSMSSLTY EDTYNTLKYASRAKKIRTTLKQNVLKSMPTEFYVKKIDEVVAENERLKERNKA LEAKATQLERAGNSGFDPLELKTWYSKIDAVYAAARQLQEHVLMGRSKIKNINY 35 RQTLKKELEEFRKLKMCVDQRVCQESF

>22:06:07|GENSCAN_predicted_CDS_7|1395_bp

	atgccttcggaacagcatcacgaatataaaagtgcggttcgctacggccgtataatgtccgtgaattggagcaaaacagcgga gttattcaaggtcattgatcgttcggcactgctgttcgatcccgacgaggaggacgatgagtccttcttcaggcgccaagcaac 40 cgtaccgcgacatcaccaagcggatgaacaaaaagtgaccatggaattcgacagggtattcgatatagacaattccaaccagga tctgttcgaggagtgacgcggcgccgctggcgcgcgggtgtaaatggatacaactgctcggtaattgtatatggagccactggcg ccggaaaaacattcaccaatgctgggcagcgaggctcatccgggtctgacctatctaccatgcaagatctcttgataagatccaa gcgcagagcgacgtgcgaagtcgatgtgggggtatctatctagaggtgtacaacgaacatgtgatgaatctgtaactaaatc gggccctttaaaacttcgcgaggacaacaatggcgtgggtgagtggtcttctgtctcacgccatctacagtcggcaggagctgc 45 taagaatgctgatgctgggcaactctcatgcactcagacccccacagatgccaatgcagagagttccaggtcacatgccatcttc caggtgcattagatcacggagcgcaagaccgacacaaaagaacgggtcaaaactatccatgatcgtatggcgggcagtgga gagggcgccagttacgaaaggcattggagtgcgattcaaggaaggcgccagcatcaacaaaagtctcttagcttgggaaattg
--	--

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cataaacaagctagccgacggcttaagcacatcccgtaccgcgactcgaacctgacacgcacccctgaaggactcgttggcg
 aaattgctgcacattgatggtggccaatgtctcgatgagtcactgacctatgaagatacctacaacacccttaagtacgtagccg
 agctaagaagatacgcacgactctgaaacagaatgtctcaagtccaagatgccaaccgagttctatgtgaagaagatcgacgag
 gtggtagccgagaacgagcgactcaaagagcgcaacaaggcgctggaggccaaggccactcagttggagcgcgcccgaat
 5 agtggttcgatccgctggagcttaagacgtggtacagcaagatagacgctgtatatgcggcccccggcagcttcaggagcac
 gtccttggtatgcgtagcaagatcaagaacatcaactaccggcagacactgaaaaaagaactggaggagttcaggaagctgatgt
 gtgtcgaccagcgagtggtccaggagagtttttaa

Drosophila Gene Hit TBLASTN with ORF2: kinesin like protein 67a (U89264)
 10 **Human Homologue** TBLASTN with ORF2: kinesin family member protein KIF3A
 (AF041853)
Drosophila EST GH22018 (AI402731)

Annotated Drosophila genome genomic segment AE003552
 15 **Annotated Drosophila genome Complete gene candidate** CG10923 Klp67a -
 motor protein

Human homologue of Complete gene candidate 2e-58 4758646 kinesin family
 20 protein 3B
 >gi|3913958|sp|O15066|KF3B
 _HUMAN KINESIN-LIKE
 PROTEIN KIF3B and also
 predicted peptide
 ENSP00000166696
 25 Gene:ENSG00000073652
 Clone:AC015936
 Contig:AC015936.00023
 6.70E-91 (predicted kinesin?:
 ENST00000166696)
 30

Putative function motor protein involved in cytoskeleton organization and
 biogenesis

35 **Confirmation by RNAi** Almost no G1 and broadened G2/M indicating arrest in
 G2/M

Example 45 (Category 4)

Line ID 442/3
Category Meiotic defects in testis: segregation defects.
Reversion ?
Map Position 70D4-7

Rescue ID H7E
Rescue Sequence

10 CGCAAGACTGTCTTCGATAGCAGAAGCGTTATTTTCGGAACATATCGTTTATCG
 AAACACTACAGTTGCTCAATACTGAACTGTCCAGCTTCGAGTAGCTGTGGCTCAA
 ACCATTGTTGTCATCGATAAGCAATTGCAATTTTATTTGTTTGCTTAAAAAATT
 AAAATATAAACTACGAGGATCAAAATATACATACATATTCCCAATATGTTAGCG
 AAAAAACATTTCTGCTCAAAAAAAGATGTTTAAATACAATGTAAGCTGTTCTA
 15 TGCATTGAACAAATTAACACATTGAGAGGTCGCTCTTATAAGTGCACATTTCA
 ATTTAAATATATTTTAAATATATTCAAATATAGTATAGCAGTATAGCATTCAAAT
 GTAAGTGTGGTTGGACTATCGCTGTAGTCCAAGAACTGCAGATAGTGTGTCATC
 GCTAGCTTTGAAGCATCTCAAAGGAAAAAGGGCGATAATTCTGATTA

20 **Genomic hit, Accession No.** CSC:AC017664

Drosophila EST CK02287 (AA141680)

Annotated Drosophila genome genomic segment AE003536
 25 **Annotated Drosophila genome Complete gene candidate** CG6650 - novel transacylase
 like

Human homologue of Complete gene candidate none

30 **Putative function** Transacylase

Confirmation by RNAi Marked increase in G1 indicating arrest in G1

Line ID 473/22
Category Meiotic defects in testis: no division (no meiosis)
Reversion R
5 **Map Position** 70A1-5

Rescue ID 2B7E
Rescue Sequence 1
10 CGCAAGACTGTCTTCGATAGCAGAAGGCGTTATTTTCGGAACATATCGTTTTAT
CGAAACTACAGTTGCTCAATACTGAACTGTCCAGCTTCGAGTAGCTGTGGCTC
AAACCATTGTTGTCATCGATAAGCAATTGCAATTTTATTTGTTTGCTTAAAAAA
TTAAATATAAACTACGAGGATCAAATATACATACATATTCCCAATATGTTAG
CGAAAAAACATTTCTGCTCAAAAAAAGATGTTTAAATACAATGTAAGCTGTTT
TATGCATTGAACAAATTAACACATTGAGAGGTCGCTCTTATAAGTGCACATTT
15 CAATTTAAATATATTTTAAATATATTCAAATATAGTATAGCAGTATAGCATTCAA
ATGTAAGTGTGGTTGGACTATCGCTGTAGTCCAAGAACTGCAGATAGTGCA
TCGCTAGCTTTGAAGCATCTCAAAGGAAAAAGGGCGATAATTCTGATAAGAA
AGTTGGCGTAGCCGGAAGGCGGATTGTCACATACAAAATAGTTTGAAAGCC
CAAACTGAG
20

Genomic hit, Accession No. CSC:AC017664
Drosophila EST LD47104 (AI515336), SD03663 (AI532240)

For other results see line 442/3
25

Line ID 670/6
Category Meiotic defects in testis: segregation defects, abnormal spindles (Ab-12/48)
30 **Reversion** ?
Map Position 70C

Rescue ID H7E
Rescue Sequence
35 CGCAAGACTGTCTTCGATAGCAGAAGGCGTTATTTTCGGAACATATCGTTTATCG
AAACTACAGTTGCTCAATACTGAACTGTCCAGCTTCGAGTAGCTGTGGCTCAA
ACCATTTGTTGTCATCGATAAGCAATTGCAATTTTATTTGTTTGCTTAAAAAATT
AAAATATAAACTACGAGGATCAAATATACATACATATTCCCAATATGTTAGCG
AAAAAACATTTCTGCTCAAAAAAAGATGTTTAAATACAATGTAAGCTGTTCTA
40 TGCATTGAACAAATTAACACATTGAGAGGTCGCTCTTATAAGTGCACATTTCA
ATTTAAATATATTTTAAATATATTCAAATATAGTATAGCAGTATAGCATTCAAAT
GTAAGTGTGGTTGGACTATCGCTGTAGTCCAAGAACTGCAGATAGTGTCATC
GCTAGCTTTGAAGCATCTCAAAGGAAAAAGGGCGATAATTCTGATTA

45 **Genomic hit, Accession No.** CSC:AC017664
Drosophila EST CK02287 (AA141680)

For other results see line 442/3

Example 46 (Category 4)

5	Line ID Category	460/20 Meiotic defects in testis: segregation defects, multipolar spindles (mitotic: High polyploids, no diploids, higher mitotic index Meiotic: Mul-02/59)
	Reversion Map Position	NR 78A1-4
10	Rescue ID	2B8E
15	Rescue Sequence	AGCTGGTCCAATTGGAAACGTTAGCTGCTCCAATGGGAGCAGCTGGCGCTCTC TCTTCGATCGCGCTCGCTCTCATCTCTCTTTAGCTTGTGCCACAGTAGCTG CCGAAGGCAATTTTCATGTGCTCGTGTGTCGACCCCCACTCAGCCCCTTCTG ATCGGAATCGGGGATTTCGGAATCGTGTAAGGCAGCCTTTGAAGGTCCCTTTTC CAGGTGGCGGCCGTATCCTTAAAGTAAACATAGTTCAACTGACTTGGCAGCGC TCCAAATGCGGTGACTTCTTGGCTATGTCATATATACCCCCACTCCCCTCCTGA CTACCCTGCCACGCCCCACCGCCACCGTCGGCGACGACAATTCATTAAAAG TTGTACGTTGTCACTTTGCGTTAACTTATCTGTGGAGCATGTTGTGCGATCGCA TTTTTATTGTCGCCATTGTCTCTCGCTCTCTCCATCGCTCTTTCGCCTGGCTTCC CTACCCTGCCCACACAGGGAAGCCTACACACTCTTAAATCATGCACTTGGAAC AAAAAGTGCAAGCATTAACTTTATTTAAACATTCAAGAGCCGCTTCTCTATT TACCATTGAAAATTTAATTTAAAATAGAAGAGGCCTTTTCAGAATAATATAAT ACCTTTAAG
25	Genomic hit, Accession No.	CSC:AC020460
30	Annotated <i>Drosophila</i> genome genomic segment	AE003592
	Annotated <i>Drosophila</i> genome Complete gene candidate	CG10588 - novel gene with homology to proteases
35	Human homologue of Complete gene candidate	2e-74 4505453 ref NP_002516.1 pNRD1 nardilysin (N-arginine dibasic convertase) >gi 2462488 emb CAA6369
40	Putative function	Novel protease
	Confirmation by RNAi	Marked increase in G1 indicating arrest in G1

Example 47 (Category 4)

Line ID 477/16
Category Meiotic defects in testis: segregation defect.
5 **Reversion** NR?
Map Position 90C5-10

Rescue ID C3E

Rescue Sequence 1

10 CTGTGGACGGTCGTCAATGCGTGAATATTCTTCTATGTGTAAGTGGTGTGCGT
GTATGTAGATTTCTGGTTAAGAAAAGCCCCAAAAACCAAAGCGCCCCGCAAA
ATATATATTGAGTCTTCTTGGCCCAACAACAAATCTGCCGCCGGACTTTCGCC
GGAGGGCGAGTGAAAAATTCAGTTTCTCTCCTCTCGACGATGCACTTTGGAGG
CTGTGTGAGTGTGTGTGCGAGTGAGTGCCTGTGTGTATACATATGCAAATGAT
15 TGGATGTGCAATCCTTGCAATCATCATCTTCATAAACTTGGCGAAAAAC
CGCAGGAAAACGCAAGCAGCCGAACAAAAAAGAGAGCCTCTCAAGACAAC
GGCAGCGGCCAAAAGTGAACGCGCAACAAACGCGGCCAAGCAGGCGCGGCA
ATTATTTATAAATCTTAAGCCGTTAGCCCCCTCTCTCTCCCACTCACGAAAAG
AAAATAAGTTAAACCAATTGGTGAAGATGATGCCCC

20

Rescue ID C3P

Rescue Sequence 2

GTCCACAGACTGGCTATATATACTAAAAACGAACTCGCGTGAGAAGACAGGG
ACAGGGCAGCAAACCTCGGTATACGAACGGAACGAAATGAAACGATTCAAGTA
25 GTAGTGTATGCAAGTCTTGTCTGTCTGCGCCTGGCGTTCTTTTCTCTCTTTTT
TCGATGGTTTTTCGCCAGGCTGGGCGCTGCCAAAACGCTGATACGGCGGCCAC
AATCACACGCGGCTAATCGCCAGTTGGGCCCTGCACAGGCTGCACATACTTTT
CACTATTAATGCGCTGTATTTCACTTATTTTTTCGAACAAATTTCGCAGCATGACG
AAGAAGCGAGCCTGTACAAGATTAGAGCGGGTAGCACGCACGATAGTATCGA
30 TACGTACGAGTATTTGGCACTGCGATACATTATCGGTGCTCGTTTCGATAGCCC
CCGATAGCTCTAGCACGAAATTTTATCGCTTTATCCATATTTTATACTATTTTT
ATTTATTGGACTTCAATGAATATTTAATTTACGTCTGGGTGCGTTTTTAAATAT
ATATGGTAATCAATAGCTGGCGAATTAGCGATATTTGAGTGTGACGCAAAAAT
GAGTTGCATCGATATCGATTTCTCGCTACTCTGGGACGCCATCTTTATTGCGG

35

Genomic hit, Accession No. AC007810

Associated ORF

40 Genscan ORF1 predicted sequences >17:48:58|GENSCAN_predicted_peptide_2|349_aa
MSRILFILLLLIVTQLSELQAAAFSVRQNRFDVDPDLQTPAPLATSTESSKKPEKAT
SGLLKKCLPCSDGIRCVPIQCPAHVRMESHEKPQICDLPAKFGYCCETGQNHT
APKPETSPKERRSGFPTILSPA VLDEARRNFEHLMHGVAQIPVRRGFPDFAHGLVF
HSTAKDDLHNFAISNSAIEQVMTTQLFGKKEQVPVEDFTNNVPIKFTETPLAHC
QPPPVCNIRSVYRSMGTCNNPEPQRS LWGAAGQPMERMLPPAYEDVPSASPA
45 AICSYTYGIASRLAPVSVVNCCTFAWQLDWTG MASGECVCVECMPAEWRLGQC
PLLHEASSEMRSLLAKS

>17:48:58|GENSCAN_predicted_CDS_2|1050_bp
 atgagtcgcattttattttttgttctacttattgtgacgcaactgagcgagttgcaggcggcagcattttctgtgcgcaaaatcggt
 ttgatgaagttcctgatttgcagactcctgcacctctggccacttccactgaatcttctaagaacccgaaaaagctaccagtggct
 5 gctgaaaaaatgccttccctgcagcgatgggtataagatgcgtgccccaaatccagtgctccgccacgttcgcatggaaagccat
 gaaaagccccaaatttgcgatctccgggtggaaaattcggtactgctgcgagactggacagaatcacactgctcccaagccg
 gagacctctccaaggagcgtcgatccggatttcccaccattctgtcacccgcagtttggatgaggcgcgtcgcaatttcgagca
 ctgatgcattggagttgcgcagattccgggtgcgccgtggcttccagatttggccatggcctggtttccactgcacggccaaggat
 gaccttcacaacttcgccatatcgaacagtgccattgaacaagtgtgaccacccagttgttgggaagaaggagcaggtgcccg
 10 tagaagatttcacccaacaatgtgcccatcaagttcactgagactccgctggcacaccattgccaaccccccagttgcggc
 aatattcggtctgtttatcgagcatggacggcacttgaataatccagaaccacagagatctctgtgggtgctgctgtgcaaccg
 atggagcgcattgctgccccgcctatgaagatttccgctcagcttctctgctgctatatgtagtatatctatggcatgcattctg
 tctggcgctgtttctgtgtcaattgttgacatttgcattggcaattgattggaccactggaatggcgagcggggagtgtgtgt
 gtggaatgtatccggcgagtggtggttggccaatgccgttgcctcatgaggcgtcgagtgaatgagccgctcttggcta
 15 aaagctag

Drosophila Gene Hit rescue sequence: eyelid/osa (AF053091)
Human Homologue BLASTX with eyelid: KIAA1235 protein (AB033061) Brain
 protein 120 (AB001895)
 20 **Drosophila EST** several including LD04852 (AA201670), LD24466

Annotated Drosophila genome genomic segment AE003718
Annotated Drosophila genome Complete gene candidate CG7467 - osa DNA binding
 25 putatively involved in DNA
 packaging

Human homologue of Complete gene candidate CG7467 - 7e-25 2588991
 dbj|BAA23269| (AB001895)
 30 B120 [Homo sapiens] and
 O14497 SWI/SNF-
 RELATED, MATRIX-
 ASSOCIATED, ACTIN-
 DEPENDENT REGULATOR
 OF
 35 CHROMATIN SUBFAMILY
 F MEMBER 1 3e-67

40 **Putative function** transcriptional regulator

Confirmation by RNAi Only wild type profiles observed

Example 48 (Category 4)

- Line ID** 496/4
Category Meiotic defects in testis: segregation defects, abnormal spindles
 5 (meiotic: Ab-08/42)
Reversion NR
Map Position 65E4-7
- Rescue ID** 2C1E
 10 **Rescue Sequence**
 GCACGATCGCTCTCTCTGGCTCTCTCTATCACTCTCTGGACTCTCTCTCAGCA
 CCTTTGCTACCGTTTTCGCAGAACAGGTGTATCGGTTTTCAAGGCAACTGTGATT
 TTTTAACCTAACATTCTATATCGAAAACCTTGTAGAGGTCGGAATTTTTCTTGAG
 CGCCTAAAAGTGTGCAGTGAAATCATTTAATCCACTTCCGGTTGCAAAACAGG
 15 AATCACACATATGAAGTGATTAAAAATCATAGAAGGTTTGACACCTTCAAATA
 ATAAGAAAACAAAATTTGTAACTGTGATAATTTATTTAATTGAAATCTTAA
 TTTAATGGCCTACAAATCTGTTGAATATCCGTTGAATACACTTTTCCAGGGTGT
 GTCCTAGTCGGCTCCTCTTTGTTACCCAGTTTGCTGGTCTTCTTAGCCGCACA
 CCAGTTTATCGCTGTTTTGCCTTTGCGCTTTTCATTATATAACAAAAACAATG
 20 TTATTGTTATTGCGGTGGCTGTAGATGTAAATGTAAATGTAGATGTAGAGGCT
 GCTTCTTGGG
- Genomic hit, Accession No.** CSC:AC018039
- 25 **Associated ORF**
 Genscan ORF1 predicted sequences >19:35:36|GENSCAN_predicted_peptide_6|190_aa
 MVSEQFNAAAEKVKSLTKRPSDDEFLQLYALFKQASVGDNDTAKPGLLDLKGKA
 KWEAWNKKQKGSSEAAQQEYITTFVEGLVAKYDNGMHKQEPNTCQARNATFR
 KSSECSLDQNTYTSSVTVIPAFHEGPKNSTASWPRIYRCYQRNQQAANCKWANTN
 30 SVCGKPHGKQSRRIFAEFLAGHTVQILG
- >19:35:36|GENSCAN_predicted_CDS_6|573_bp
 atggtttccgagcaattcaacgccgccgagaggtgaagagcctgaccaagcgtccagtgatgacgagttcctgcagctg
 tacgccctgttcaagcaggccagcgttggtgacaacgacaccgccaagccgggtctcctggacctgaagggaaggccaagtg
 35 ggaggcctggaacaagcagaagggaagagcagcaggccgccagcaggagtacatcacctttgtggaggcctgtgtgc
 caagtatgacaatggaatgcacaacaagaaccaaacacttgccaagcagcaatgcgactcggtttcgaaaagctcggaatg
 ctcgctggatcagaatactatagtcagtggtgtacggttatctgcattccacgaaggccaagaactgcagggcaagttggc
 caagaatttaccggtgctatcagcggaaaccaaacgagcccaactgcaagtgggcaaacacaaatagcgtttcggggaaccc
 cagggaaaacagagccgccgaatcatttcgcagaatttcggccggccatacgggtgcagattcttgggtaa
 40
- Drosophila Gene Hit** rescue sequence: melt (S144114) P element insertion site
 (AF174669), TBLASTN with ORF1: diazepam binding inhibitor
 (DBI) (U04823) and melted (AF205831)
- 45 **Annotated Drosophila genome genomic segment** AE003560
Annotated Drosophila genome Complete gene candidate CG8624 melt - putative signal

156

		transduction protein CG8631 msl-3 - acyl-CoA- binding protein/diazepam binding inhibitor
5	Human homologue of Complete gene candidate	CG8624- predicted gene ENSP00000065899 Gene:ENSG00000055889 Clone:AC015904
10		Contig:AC015904.00014 1.70E-15 (unknown predicted gene 1: ENST00000065899 and AK022666 Homo sapiens cDNA FLJ12604 fis 2e-29
15		CG8631- gi5803104 0C85AE40FDF874CD [refNP_006791.1 male- specific lethal-3 (Drosophila)- like 1 [Homo sapiens] (1.70E- 36) and Ensembl predicted peptide ENSP0000006617 Gene:ENSG0000005302 Clone:AC004554
20		Contig:AC004554.00001 8.70E-19 (unknown predicted gene 1: ENST0000006617
25		
30	Putative function	CG8624: putative signal transduction protein CG8631:acyl-CoA-binding protein/diazepam binding inhibitor
35	Confirmation by RNAi	CG8624: reduced G1 and G2/M Indicating fewer cycling cells, CG8631: Increased G1 to G2/M ratio indicating arrest in G1

Example 49 (Category 4)

5	Line ID Category	523/19 Female sterile. Meiotic defects in testis: cytokinesis defects, segregation defects (Mitotic: Less condensed chromosomes, nuclear bridges, Meiotic: Seg-01/02)
	Reversion Map Position	R 75C1-4
10	Rescue ID	2B4E
15	Rescue Sequence	ACTGAGAGCATATTTGTGCACCAGAGGGCTGCATAACAACATTCTCTTTGTCC ATTCGTTATACTTCGTATTCAGAATACATGTCATTGAGTTGGTCCCGTTCTTTTT GCGTTCACCTTCGTATATATTCGGCGATCGAAATGAACTAACTGAATGTGTTCA AAGAATGAATGAAGCCAATGAATTTTCAATAGTAATTCAGAGTGCTTAAAATT CTTTCATGTTGTCATTGAGTAAAATGAGTTCGGACAGCGCGAAGGTAAGTCGAA GTTTGTGTTTTATTATGTTTTATTTGTATTATTATGTACACTAGTCGGCATACTTT TGCGTGCGTCTTATACGTGTGCGTCTTATTTAACAATATTGTAAAATAAAATAT ATAAATTATTTGTTATATGCGTAGGGGCCTTTATTTTGTGTATTGATAGTCTTTT 20 GTCATAGATATCATTATTCTGACAAGATTTGAACTTTTCAAGTTATTGCCTCTC GTTATTCAATTCTAGCTGGTCTTACGTTACGCGATATTTCTAAAATATCCTA AAATCGCACAAAACAGTCACGCCACACTTTTGAAAAACGTGGTAATATTTTT CATACTTGCATTAAGTCTGG
25	Genomic hit, Accession No.	AC007691
	Annotated <i>Drosophila</i> genome genomic segment	AE003520
	Annotated <i>Drosophila</i> genome Complete gene candidate	CG4306 – novel
30	Human homologue of Complete gene candidate	4e-25 3242764 (AC005154) similar to protein U28928 (PID:g861306) [Homo sapiens]
35	Putative function	No homologies to indicate function
	Confirmation by RNAi	Only wild type profile observed

Example 50 (Category 4)

Line ID 666/19
Category Mitotic defects in brain: anaphase defects
 5 (weak, overcondensation, aneuploidy, lagging chromosomes, metaphase with bipolar spindle)
Reversion NR
Map Position 64E1-5

10 **Rescue ID** I9E
Rescue Sequence
 CCCTCGTCTACGTGCGAAATTCTGGATGCTTCTCGGATTTAGGGTTGTATCTCGA
 AAACGTTTGACTGCGAATGTCAATATCGATATGCTAACCGATAGCTGTCGATG
 TTTTAAACACAGTGCAGTGTGTTTAAATCGCTCCCATTTATATATATTTGTGC
 15 NTGCTTTTGGCGGTNNNTTTCTTTATATGCTTCTTATGCTTTTACGATTATTATT
 AGCGCTTATTTGATTGCAAATGCCAAGGAAAGCGTGACTGTGATGGCGAAATG
 CGGAAAGTACTCCTTAAATCTCATATATCGCATAAACTATCGGTTCTGGAAT
 GTTTCGTGTAAGTCTGCGAAGATAGAGATCGATCTATTTTGAGGATACATTTG
 TTAATATTATAAGGGATTCTTCTACAGGGGTCAGATTGCTTAAAAACACACAG
 20 AANAATAAACAAAATATTTCTTTGAAATATTGAAATATTTGAAATANAAAAAA
 CGTATTGACGAGGTAAGCATATTGAAAAAGATAGGAAGGTGATGGAGAAAGT
 GCACTTATATTGGTCACCAAAGAGCTTATAATCAAAAGATCAATAGATATAAA
 TATCTTTATATGATATAAAATATAATACATATAATATAATATCATATACAATG
 GATAAATTGCAAGTGGCAAAATGAATTCGCGGAATTAATTCTGAANCGAAA
 25 GGGCCT

Genomic hit, Accession No. CSC:AC014815

Associated ORF

30 Genscan ORF1 predicted sequences >17:46:43|GENSCAN_predicted_peptide_1|334_aa
 MGKDFYKILGLERKASDDEIKKAYRKLALKYHPDKNKSPQAEERFKEIAEAYEVL
 SDKKKRDI FDNYGEDGLKGGQPGPDGGGQPGAYTYQFHGDPRATFAQFFGSSDP
 FGAFFTGGDNMFSGGQGGNTNEIFWNIGDDMFNAQAPSRKRQQDPPIEHDLF
 VSLEEVDKGCICKMKISRMATGSNGPYKEEKVLRITVKPGWKAGTKITFPQEGDS
 35 APNKTPADIVFIIRDKPHSLFKREGIDLYTAQISLKQALCGALVSVPTLQGSRIQV
 NPNHEIIPKPTTTRINGLGLPVPKEPSRRGDLIVSFDIKFPDTLAPSLQNQLSELLPN

>17:46:43|GENSCAN_predicted_CDS_1|1005_bp

40 atgggcaagacttctacaagattctgggctcgagcgaaggccagcgacgatgagatcaagaaggcctaccgcaaaactggc
 actcaaataccatcccgacaagaacaagagcccacaggcggaggagcgcttcaaggagatcgccaggcgctacgaggtgctg
 tcggacaaaagaagcgcgacatcttcgacaattacgggtgaggatggattgaaggcgagacagccgggaccagatggcggcg
 gtcagccgggagcgtacacttaccagttccacggcgatccgaggggccacatttgccagttcttggatcgctggatccggttggc
 gcgttcttaccggcgcgataacatgttagtggcggtcaggggcggaataccaacgagatcttctggaacattggcggcgacg
 atatgttgcctttaatgccaggcaccagtcgcaagcgccagcaggatccgcccacgagcatgatctgttcgtgctgctggag
 45 gaagtggacaaggggatgatcaagaagatgaaatctcacgcatggccaccggaagcaatgggcccgtacaaggaggagaag
 gtgctgaggatcacagtgaagccgggctggaaggccggtaccaagattaccttcccccaagggtgattcggcgccaaaca

gacgccagctgacatcgtcttcattcgcgacaaaccgcattcgtgttcaaacgcgaggggaatcgatctaaagtatacagccc
 agatcagctctgaagcaggccttggtgcggagcactggtagtgcccacgctgcagggcagcaggatacaggtgaatccgaacc
 acgagatcatcaagcccaccacaacgcgcggatcaacggactgggtctgccggtgcccaaggagccatcgaggcgcggcg
 atctgacgtctccttcgacattaagtgtcccgacacactggcaccagctctgcagaatcagctgtccgagctgtgcccaactag

5

Drosophila Gene Hit rescue sequence: fasciclin I (FasI) (M32311) TBLASTN with
 ORF1: DnaJ homolog (DROJ1) (U34904)

Human Homologue TBLASTN with ORF1: DnaJ-like heat shock protein 40 (HLJ1)
 (U40992.2)

10

Annotated Drosophila genome genomic segment AE003565

Annotated Drosophila genome Complete gene candidate CG10578 - DnaJ-1 a
 chaperone putatively involved
 in protein folding. Stimulates
 activity of HSP70

15

Human homologue of Complete gene candidate 8e-94 1706473 P25685
 DNJ1_HUMAN DNAJ
 PROTEIN HOMOLOG 1
 (HDJ-1) (HEAT SHOCK
 PROTEIN 40) (HSP40)

20

Putative function Chaperone involved in protein folding

25

Confirmation by RNAi Almost no G1 peak, increased G2/M indicating G2/M arrest

Example 51 (Category 4)

Line ID 714/11
Category Meiotic defects in testis: cytokinesis defects, abnormal spindles
5 (Ab-01/04)
Reversion ?
Map Position 66A10-15

Rescue ID 2A4E
10 **Rescue Sequence**
AACCAGAACGAAACTCCAATGCAGTTTCATTTTGTGAGTTTAATCATTAAACA
AAGAACTGCGCAACCGATCGCAACTAGCTCGTGGACTCTTGTTCTCCCAATAA
TTGGTATGTTTTCCATTTTGCCTTAACATGGAAAATGTGTGAAAAGCTTTTTCC
CCCTCCAAAAGAAGCGTACTGAACTAAGCTTTCGGTGGTAGTAATAGTAGTC
15 GTTATATCTTATTTTTCTTATTACGTGCAGCTGCAATCATTGGCTGCGTCACTT
TGGCGTCAGCTATAAACTGGTGGATCAACTCGGCGGCCTCCAAAAGCTGCGCA
TCTGCTCCAGACACTTTAGCCAACGCCAGGAGATGGCCAAAACCCGCATCAA
GATGACGCCGCTGCGCAAGTCCTCGTCTCCAAGGGCATTGTGCTACCCATTA
ATGCCGCTGGAGGGTCGGTCATTGCAGGCGCCTTAGCACGAGGAGGAGGTGC
20 A

Genomic hit, Accession No. AC012390

Associated ORF
25 Genscan ORF1 predicted sequences >19:47:45|GENSCAN_predicted_peptide_2|711_aa
MRSHQAVGNLLLADEALPAVQSASVYVWMAEQPLSPGQSYDIKIADSPSVSS
KSITDNGADVQWFAFEHSQYYQGVQQMFLSALERIDSEFLITLIKRCPHYVDSLQ
LSEVCKMTEDFSLASELLERALLLLESSLHINFSLTSGNCRDLYRRQENRSFYVLF
KHAQYLEERACSRFAFEISKLLLSLPDTPDLAMILPNQPDQCTGNMTQLQQAGK
30 IRKRSEKQFPIGTEPRGTDALRFTLQTLASAGRDITWNIKRLQGSRTVGAQGYLI
DKKTA VQYKITIAHLKDPNIDQLFDSSGDGKADLHGSTPDWGCQAMMADAISR
YKEGNPVFYTWTPYVWSNELKPGKDVVWLQVPFSALPGDKNADTKLPNAGGI
EGLIADEEVQVLDALCDAPCVGVSHSCRLDGNRRGNRELRLFPKGSQFGVADG
CADKQSVMEYHAAKTGHTKFSESEEEKKALTEEEKKAQLALIEEKLKQKRIREE
35 REKIEALQREKNRIKSGKDMTEAKRRMEELEMKKIVEQRKREKDEEKAARDRVK
AQIEADKAARKAREQKELGNAEPAPSVSSTTVSSPPAGVKSPPRDYETETRIQGASA
ILAAAAPYYQPPAVPQDVQPDRIPIGYAFGVVCGSHISGWHCSAGHYEDGNENFE
CLKTFSTSDRIGCEWRWAAATVLAATCISPNGRCGHYKRVRRRIKTNITTT

40 >19:47:45|GENSCAN_predicted_CDS_2|2136_bp
atgagatcgatcaagccgttgcaatctgctgctggcggcagacgaagcgtaccggcgggtgcagagcgcgtggtgatgtg
gtatggatggcggaaacgcgcttctccaggcagagttacgacatcaaaattgccgactctccatcggtgtcctccaagtctatc
acagataatggagcggacgttcaatggttgcttggagcatagccaatactaccaggagtgagcgaatgttcttctgctctg
agcgcattgactcggaaattctgatcacacttatcaaacgtgcccctatcatgtcgactccttggttcaactcagcgaagtatgcaa
45 gatgaccgaagacttttcttggtcctccgaactgcttgagcgcgccttctccttctggaatcgctgctgacatcaactcagttga
cgtcgggcaactgccgactggactaccggagacaggaaaaccgatccttctacatcgctgctgttcaagcagcgcagttacgtg

aggaacgagcttgagccgcaccgcttcgagatctccaaactgctcctgagttcagccagacacagatcctcttgcctgatt
 ctaccaaatacagccggatcaatgtaccggcaatatgacgcagctgcagcaggcgggcaaaatccgtaagcgctcagaaaagca
 gtttccgactgggtactgaaccgcgggtactgacggttgcgttcaccctgcagacactggcgtctccggcgacatcacct
 5 ggaatataaagcgtctgcaaggttcccgtgttaccggcgccggccagggttacctcatcgataagaaaaccgctccagtaaa
 aatcacatcatcgctcatctgaaagatccgaatatcgaccaactgttcgattcaagcggcgacggaaaagcggtttacacggta
 gtaccccgactggggtgccaagctatgatggccgacgccatcagtcgtacaagaggggcaacccgggtttttatcacctg
 gacgccgtactgggtgagtaacgaactgaagccgggcaaatgagtcgtctggttcaggtgcccgttctccgactgccgggcca
 taaaaacgccgataccaaactgccgaatgccggtggcatcgaaggcctcatcgccgatgaagaagtcaggctctcgtatgcct
 10 ttgtgatgcggcgtgtgttgggtgtctccactcgtgcccagctccttgatggcaatcgccgagggaataatgaactgcggcttttatt
 cccggcaaatcccagtttgagtagctgatggatgtgcagacaagcagagtgttatggagtaccatgccgcaaaaccgggtcac
 accaaattctccgaatcggaggaggaaaagaaggcgctcaccgaggaggagaagaaggccagctggccctcatcgaggag
 aagctcaagcagaacgcacgcgagggagcgcgagaaaatcgaagccctgcagcgggaaaagaatcgatcaagtcc
 ggcaaggcatgaccgaggccaagcggcgcatggaggagttggagatgaagaagatcgttgagcagcgcaagcgcgaaaa
 ggacgaggagaagggcgcccgcatcgggtaaggctcaaatgaggcggacaaggcagcacgcaaggctagagaacaaa
 15 aggaattgggcaacgcagagccagctccatccgtgagctccaccagtttcgtcaccaccggccggtgtgaaatctccgccgc
 gagactacaccgaacccgcatccaggcgccagcgcaatcttggccgcagcggctccctactatcaaccggcggtgttccc
 caggatgttcagccggatcgtcctatcggtatggagcattcggagttgtctgcgggtcccatcagcgggtggcattgttctgcg
 gggcattatgaagatgtaataaaaatttcgagtgctcaagacatttcgacttctgaccgcattggctgcgaatggagatggcg
 gcagcaactgttcttgcgcaacctgcattagcccgaacggcgttgcgggcattataaacgcgtacgtcgtcgcattaaaacaaa
 20 cataacaactacgtga

- Drosophila* Gene Hit** rescue sequence and BLASTX with EST: BIP1 (Y14998),
 BLASTX with genomic sequence matches BIP.
Human Homologue BLASTX with BIP1: alanine:glyoxylate aminotransferase
 25 (X53414) ?
***Drosophila* EST** GM04749 (AA695904), GM13608 (AA803601)
- Annotated *Drosophila* genome genomic segment** AE003556
 30 **Annotated *Drosophila* genome Complete gene candidate** CG7574 - bip1 unknown
 function
 CG13681 - unknown
- Human homologue of Complete gene candidate** none
 35
- Putative function** no homologies to indicate functions, *Drosophila* Bip1 interacts with
 transcriptional activator Bric-a-brac which is required for ovariole
 formation
- 40 **Confirmation by RNAi** Both show reduction in G1 and G2/M indicating fewer
 cycling cells

Example 52 (Category 4)

	Line ID	763/4
	Category	Meiotic defects in testis: segregation defects (overcondensation, fewer anaphases)
5	Reversion	R
	Map Position	90F
	Rescue ID	2F5E-1
10	Rescue Sequence	CGGCAATGTCTGCGCCCCCAATCTGAACTTGCCTCGCCCTCTCCGCCCCTGATC TCATCTCCTCTTCAAACCCCTGCTCCCCTTTTCTGCACACATTAACGTCAGCCT TTAAGTGTGCTTTCTCAGGTGCTGCCCCCTGCGCCACCATCCCCCGCTCCATG CTCTTTCCATCTTGCGCTCTCTGCGTTCTATCTACATTTTTTTTCGAGGTCGCGCG 15 CTGCTTTTTCCGTTGATGTTTCGTTCTCGTCAATGTCGCAATATGCGCAAAAGGC AGACAAAAAAAATGAGTGGAAAAAGTACATACATACCGGTGATTGATGGG CGGTGGGTGGCGGTGGTGTAGGNGTGGTTTG
	Genomic hit, Accession No.	AC006495
20	Associated ORF	Genscan ORF1 predicted sequences >22:47:02 GENSCAN_predicted_peptide_3 283_aa MTERENNVYKAKLAEQAERYDEMVEAMKKVASMDVELTVEERNLLSVA YKNVI GARRASWRIITSIEQKEENKGAEKLEMIKTYRGQVEKELRDICSDILNVLEKHLIP 25 CATSGESKVFY YKMKGDYHRYLAEFATGSDRKDA AENSLIA YKAASDIAMNDLP PTHPIRLGLALNFSVFY YEILNSPDRACRLAKAAFDDAIAELDTLSEESYKDSTLIM QLLRDNLTLWTSMDQAEIPIPKLPDRQSKTTLIFSPRSQVNP KILHKNN TI GRVIC SVFA
30	>22:47:02 GENSCAN_predicted_CDS_3 852_bp	atgactgagcgcgagaacaatgtgtacaaggcaagctggccgaacaggccgagcgctacgacgaaatggaggccatga agaaggtcgcctccatggacgtagagctgaccgtcaggagcgaaatctgctgctggcggtacagaatgtgattggagcac gccgtgcctcgtggcgcacatcacctcgtcgaacagaaggaggagaacaagggggccgaggagaaattggagatgatcaa aacctaccgcggacaggtggagaaggagctgcgcgacatctgctcggatatactgaacgtgctcgagaagcatctcattccatg 35 cgccacatccggcgaaagcaagatttactataagatgaaggcgactaccatcgctacctggccgaattcgccaccggctcc gaccgcaaggatgcggcagagaactcgtgattgctacaaggcgccagcgatattgccatgaacgatctgccaccaacaca ccccatccgttgggcttgccattgaactctcgtgttctactatgagattctcaactcgccggaccgcgttgccgcttgccgaaa gcccgttctgatgatgccattgccgagttggatactgagcgagagagctacaaagactcgacactcatcatgcagctgctgc gggacaacctcaccattatggacgtccgatatgcaggcagaagagattccgattccaaaactccccgacagacagtcacaaacca 40 cattgatttttagccccgaagtcaagtaaacccaaagattctccacaagaacaacaccatcatcggcagagttatctgtagcgtgtt tgcgta
	Drosophila Gene Hit	rescue sequence: 14-3-3 epsilon isoform gene (U84898) TBLASTN with ORF1: 14-3-3.
45	Human Homologue	TBLASTN with ORF1 and BLASTX with 14-3-3: epsilon isoform 14-3-3 protein (U43430.1)

	Annotated <i>Drosophila</i> genome genomic segment	AE003721
	Annotated <i>Drosophila</i> genome Complete gene candidate	CG8045 complex gene
5		appears to encode 3 things : Transcript: CT24102 unknown Transcript CT24072: transcription factor RNA polymerase II transcription factor ,
10		Transcript: CT24092: diacylglycerol- activated/phospholipid dependent protein kinase C inhibitor /14-3-3 protein epsilon (suppressor of ras)
15	Human homologue of Complete gene candidate	CT24092: e-119 NP_006752.1 tyrosine 3- monooxygenase/tryptophan 5- monooxygenase activation protein, epsilon polypeptide; 14-3-3 epsilon [Homo sapiens
20		
25	Putative function	transcription factor, or 14-3-3 proteins which associate with cdc25 phosphatases
	Confirmation by RNAi	CT24102: wild type profile only, CT24072: Loss of G1 peak CT24092: Increase of G1 peak

Example 53 (Category 4)

	Line ID	951/8
	Category	Mitotic defects in brain: (some overcondensation, anaphase bridge, metaphase with swollen chromosome and bipolar spindle)
5	Reversion	NR
	Map Position	73D
10	Rescue ID	2E8S
	Rescue Sequence	
	GTATAAACAAGATCCCGAGACACCGGTCAGTTGGTGCTACACGCTCTTGGAGA	
	GCGCTGTGTTTGTTCGGTTCAGCGATTAGCGATAGTTTGTTCGAGCCGGTTGT	
	GTTAACTTGGCTAGCTTCGGGTTTATTGTGACACTTTCCCAAATCGATCGTTT	
15	GCGAAGCGTGTCATAGCGGAACATACATACATAGATAACCAGCGTGTCTGGGT	
	GTTTCATGAAAAAGAGTGCGTGATATGGGATTCGATATGGCAACACGCTTTATG	
	GATATACTAAAGCTGACCTTTAAGTGAGTTTTCCCAGTCAGTGTCCGCTTCTTG	
	CTCTTGCGGAGCGTTAAACGGTTTTCTGTGTTTTGAGGTCTCGCGTCTTGGTTT	
	TGCAACAGCTTCTGCCCAGCATGCACACATACGTGTGCACTGGGAAAATAGTG	
20	TTGCAGAAAGTGCTTGATTATAAAATATTACAAAAAATGTGATGAAACACTTTT	
	TATTTTCTTCAAAAAATCAAGATAAATTAACACTATCCTGCTCTTAAACAT	
	GGAGATTAATTCAATTTTAATTAATAAATAATTTTTTTTACAATTTATGATTTA	
	TGAATTTATGCACTCCTTGAAACTATTAAAGACTCAACAGTGA	
25	Genomic hit, Accession No. CSC:AC015272	
	Associated ORF	
	Genscan ORF1 predicted sequences	
	>23:03:05 GENSCAN_predicted_peptide_1 602_aaMGFDMATRFMDILKLTfKPFKTN	
30	YTEEKYFNDKLRSSKNIERRYILDVGFRGPTAVTYNPIWVISFKYEQRKLSTAIYSV	
	IKTKSGPVRGVKRNTIWGGSYFSFEKIPFAKPPVGDLRFKAPEAVEPQDQELDCTS	
	PADKPLQTHMFFRKYAGSEDCLYLNVIYVKDLQPDKLRPVMVWYGGGYQVGEA	
	SRGLDVVIVTVAYRLGALGFLSLDDPQLNVPNGAGLKDQIMALRWVQQNIEAFG	
	GDSNNITLFGESAGGASTHFLALSPQTEGLIHKAIVMMSGVLCPTWQPPRNNWAY	
35	RLAQLKGYTGDNDKKAIFEFLRSMSSGGEIVKATATVLSNDEKHHRLFAFGPVVE	
	PYTTEHTVVAKQPHELMQNSWSHRIPMMFGGTSFEGLLFYPEVSRRPATLDEVGN	
	CKNLLPSDLGLNLDPKLRENYGLQLKKA YFGDEPCNQANMMKFLELCSYREFW	
	HPIYRAALNRVRQSSAPTYLYRFDHDSKLCNAIRIVLCGHQMRGVCHGDDLCYIF	
	HSMLSHQSAPDSPEHKVITGMVDVWTSFAAHGDPNCESIKSLKFAPIENVTNFKC	
40	LNIGDQFEVMALPELQKIEPVWNSFYAPNKL	
	>23:03:05 GENSCAN_predicted_CDS_1 1809_bp	
	atgggattcgatattggaacacgcttatggatatactaaagctgacctttaagccatttaaagcgaactacactgaagaaaagtattt	
	caatgacaaactcagatcttcgaaaaatattgaaaggcgttatcttgatgttgcttcgcggacccacagcagtcacgtacaat	
45	ccaatctgggtaataagcttcaagtacgagcagcgcaaatgtcaacagcaatatattcgtcataagacgaaatcaggtcctgtg	
	cggggagtggaagagaacacaatctggggagggaagctacttcagtttcgagaagatacccttcgaaagcctccggtgggagat	

ctgcgcttcaaggccccggaagcagtggagccatgggagcaggaattggattgcacttcgcccgcagacaagccccctcagaca
 cacatgttttcagaaaatacgcgggctcagaggactgcctctactaaatgtgtatgtcaaagatctgcagccggataaactgcgtc
 ccgtgatgggttgatctacggaggaggctatcaagtggcgaagcttctcaggattggatgtgtcatagtcaccgttgcttatcg
 actgggtgccttgggttcctcagcctggatgatccccaaactaaacgttcccggaaatgcaggtctcaaggatcaaatcatggccc
 5 tgcgatgggtgcaacaaaacatcgaagcattcggcgggtgattccaacaatattacactctttggcgaagtgcggcgaggcctc
 gaccacttccttgactaagtcaccaaaactgaaggtcttatccacaagctatcggtatgtcgggcagtggtttgtgcccctggacg
 caaccaccgagaaataattgggcttataggctggcccaaaatgggatacaccgggtgacaataaggacaaggcgatctttgagt
 ttctgcgatcaatgagtgggcgggagattgtcaaggccaccgcaacagttctcagcaacgatgaaaagcatcatcggtaccccttc
 gccttcggacctgtcgtagaacctatactaccgagcacactgtggtcgctaaacaaccgcatgaactgatgcagaatagctgga
 10 gtcacaggatacccatgatgtttggaggcacgagcttcgagggattgtattctatccagaggtttcaaggcggccagcaaccctc
 gatgaggtgggttaactgcaagaatctgctaccgagcgatctcggtcttaacctagatcccaactgcgtgagaactacggcttgca
 actgaagaaggcgtatttcggcgacgaacctgttaaccaggcaaacatgatgaagtttctcagctatgctcatatcgagagtctg
 gcacctatatacagggcagctttgaaccgtgtccggcaatccagcgcacccacgtatctgtatcgattcgatcacgattccaaact
 gtgcaacgccattagattgtactttgcggccatcagatgcgaggtgtttgtcatggtgacgatctgtgctatatttccacagcatgtt
 15 gtcgcatcaatccgctccgattctccggaacacaaggttataaccgggaatggtcgacgtttggacgagtttcgagcccacgga
 gatcccaactgcgaaagtataaaatcactcaagttgcacccatcgaaaacgtaaccaactttaagtgctcaatattggggatcagt
 ttgaagtcatggcgcttcagaaattgcagaaaatcgaacctgtgtggaatagtttctacgccccaaacaaactgtag

20 ***Drosophila* Gene Hit** TBLASTN with ORF1: alpha esterase (aE10) gene (U51054)
Human Homologue TBLASTN with ORF1 and BLASTX with U51054: bile salt-
 dependent lipase (S79774)

25 **Annotated *Drosophila* genome genomic segment** AE003671
Annotated *Drosophila* genome Complete gene candidate CG1131 - alpha esterase 10
Human homologue of Complete gene candidate 4e-48 4557239
 ref|NP_000656.1|pACHE|
 acetylcholinesterase (YT
 blood group) precursor
 30 >gi|113037|s

35 **Putative function** alpha esterase
Confirmation by RNAi Only wild type profiles observed

CATEGORY 5: SMALL IMAGINAL DISCS (BLOCK TO PROLIFERATION)**Example 54 (Category 5)**

	Line ID	113/20
5	Category	2nd chromosome, small imaginal discs
	Reversion	R
	Map Position	50D/E
	Rescue ID	EcoR1
10	Rescue Sequence 1	
	CTGAGGCNCTTTGCCAATATGTGTATATTGGGCGGGGNACATGCGTNAATCGG	
	TTAAAGCCGCTACTTACATTCTGTTCTTTGCATCTCCCCATCCACAGCTATAA	
	AGCAAGATGAGCTACGCCGCTGATGTGCTGAACTCGGCCCATTTGGGAGCTCC	
	ATGGTGGTGGCGACGCCGAGTTGCGTCGTCATTTCGATCCACGGNCCATGAT	
15	TTGGATGCATCCTTCGCCTTACACGCTTCGCANATCTAAAGGGGCGCGGCTG	
	CAAAGTGCCCGCAAGGAAGTNGCTCCCCACCT	
	Rescue ID	BamH1
	Rescue Sequence 2	
20	CCACCTGGTACCACAGCGCTCANACGTGTATGTACACGGATTTTCTGCCGCGT	
	GTGTGTAGCGCGGCCCGTGATTGGCTGCAGTCGCGATGGCGGGCTAAAACGGG	
	CGAAGTCAGTATTTCTCCCTGTGACGANGCGAGCAACGTGAACAATGCCCAC	
	TCATTTCAATTGCAAAATGCCAAAAGTGCGCGCTTTGAATTGGCCATTTGGT	
	TCGTTGCGTTTCGTTTGTCTTTTGGTACTTACGTTTGCTTGTGCGATTGTACAAA	
25	GATAATTGTAGAGTAACGTTAGCAAATTATATTTATTTTGCGCCTGGTTTTTGC	
	TTTTCCAACGANCGAGATGTCAACAGGGTTGTATTANCGTGTGCGGCTGAT	
	TCGATATTTGGGATGCCGATTGTCTGAAGCGANGGTTCAACGGGGCTGCCAAC	
	TCCCCGAAAATCTATCNATGGTATTGTGCGCCAAGGGTAAAAATAAATAAAAA	
	TATGTTAAAACCGCGGAATAAATGGGGGAACCGAAGTGGAAGTGTGGTTCA	
30	CAGTGCTCTGACTTTCGGGAGCAGTTAATATAGTTGGCATTAAATTCAATTAGA	
	GCTCCAAAGTGCTGGTCACAAAGAACGCACAAGAACGGGCCATGAAAAACCT	
	GTTGCGCCAGCAGAACGAAAAGTAAAAATTAGAAGAAACCAT	
	Genomic hit, Accession No.	CSC:AC017131
35	Drosophila Gene Hit	rescue sequence: selenophosphate synthetase (ptuf1) (U91994)
	Human Homologue	BLASTX with U91994: SELENIDE, WATER DIKINASE 1 (SELENOPHOSPHATE SYNTHETASE 1) (SELENIUM DONOR PROTEIN 1) (P49903)
	Drosophila EST	LD46437 (AI514756 similar by BLASTN to U91994
40	selenophosphate synthetase (ptuf1) gene)	

Annotated *Drosophila* genome Complete gene candidate CG8553 selD selenophosphate synthetase

Human homologue of Complete gene candidate

1711372 P49903

SELD_HUMAN

SELENIDE, WATER

DIKINASE

(SELENOPHOSPHATE

SYNTHETASE (1e-159)

5

10

Putative function selenophosphate synthetase

Confirmation by RNAi Only wild type profiles were observed

Example 55 (Category 5)

	Line ID	121/1
5	Category	2nd chromosome, small imaginal discs
	Reversion	NR
	Map Position	60B
	Rescue ID	BamH1
10	Rescue Sequence	
	TCCTGTGCACTCATATTGATTTGCCTTGTCAAGTGGCTAAAGAAATATTTAAATG	
	TTTGTTATTTCTGTTGCTAGCGCTCCGACAGTCTGGCAGCACTGCTCGCTGTCG	
	ATAGTTCAACTGAGTTGCTGTTTCATCGAACAGAGCTGCCAACTCTATTTTGT	
	AGCTGGCCAGCCAGGATTGCCAGAGTAAGGCCCTCAAATCAGCTGTTTTGTGT	
15	TTTGATTTTATTTTGAAAGTCCTAGTTTAAAATTATGCTTTCTCCGACAGATCA	
	GCACAAATAATACTAATAAAGCTCACAATGCTAAGGTTGTGCCTTCCAACTCG	
	AGCTGGATATGTGCGTAAGTAAGGACTTTACGTCTATAAACTTGTTATGTAA	
	AGTAAATGTTTGCCTATTGCGCAATTTCTCCAACGAAAAACCCAGAAAACCNA	
	AACCCCTTNAANTTTGGAATATNCCCAATGAATGCAGCACCCGTGAAATCC	
20	GTAATGCCTTTGTCCAGCTCTCCAAATTGGTAAGTAAGTCAAGATCCAAAGG	
	AGCCTCCTAAACCCTGCCCTTTCCACAGTACCACCCAGATGTTAAGAGCAATG	
	CTGCGTGTCCGGAGCGCACAGCCCGATTTGTTTCAGATCTCCGAGGCGTACAAG	
	AACCTGATAAAGCCGGAACGGAAGGAAAAA	
25	Genomic hit, Accession No. CSC:AC020499	
	<i>Drosophila</i> Gene Hit rescue sequence: DnaJ60 gene for dnaJ-like protein (Y11900)	
	Annotated <i>Drosophila</i> genome genomic segment	AE003463
30	Annotated <i>Drosophila</i> genome Complete gene candidate	CG12240 – DnaJ60
		CG13570 – spaghetti ser/thr phosphatase
	Human homologue of Complete gene candidate	CG12240- 4827026
35		ref NP_005138.1 pTID1 tumorous imaginal discs (<i>Drosophila</i>) homolog >gi 3372677 (AF061749) 7e- 08
40		CG1116- 2495728 HYPOTHETICAL PROTEIN KIAA0258(aa)
45	Putative function	CG12240 : Chaperone involved in protein folding CG13570 : serine/threonine phosphatase

Confirmation by RNAi

CG12240: Marked reduction in G1 and G2/M peaks
indicating fewer cycling cells
CG13570: Marked increase in G1 peak

Example 56 (Category 5)

Line ID 127/2
Category 2nd chromosome, small imaginal discs
Reversion NR
Map Position 57F

Rescue ID EcoR1
Rescue Sequence 1

10 GCCGGTGGGCCCACACTTGTNCGCCCGCGCATCGGCTGTCTGTGGGAGTGCGA
NCGAGTCAGATAGTAGATCCGATGCGCTCTCCAGATACTTTTTGAACACTGAA
GAAAACGCGCAGTTGTGGGTGAATTCAGCATCATCAGATTGAATCACACACA
ATCCTAGTCGCCTCACGCGAAGAGAACTATGTCATGATCAGATATCGGTGTAT
GCATTCTATATTATGTACTTCGAAATATGTAATTTATTAAGTTTTCGCTATACT
15 TTTCATTCAAATTGGCAAAAACCAATTCAAAGGTTTTCAATATTTTCGAAAAG
CATTTTAGGCTTTCTATGTAACGTATGTTTTTCAAACAAAATATTAGTTTTTGA
AACTTTATTATCGGATAAAACAAATGTAAGCCAAATNACAACGTNTATGATAC
TCCCAAAGATCCGCNCTNTTAAAGTGGCCTAAAAATAGCTGACGCATTAANCC
ATAGGCGCTTCGCTTCTCAAGATAAAACCTGGCGTGCTCAACTCAAGAAACAA
20 ATATGTGGTTATATACATATATACATATATGGGGCATATAACCGATGTGTGAC
GTGACATTGGCTCGTTCTATTACATACTTAAACACTAAATGCAAACCTATCA
AAAACCNACTACACTAAGCGAAAAACGGCAGANATAGTTAAGGAAAGTGGTC
CA

25 **Rescue ID** BamH1
Rescue Sequence 2

CTTCTTTTCTCAAAAAACGTCGCTCGNGTCCCNCAATCGTTTTACAAACTTCGC
TCGGAACGGACGTGTGCGCGCTCTGAAAGGAAAAAGTGAAAAAGTGTGTGAC
AAAGTGCAAATAAGCCACAACGCGCATGTGAGAAATCAAATTTAATTGAGAA
30 GCATCAAAAATTGTATACATATCGAGCGTATCCACATCGCTGTATGTGTGAGT
GTGCCAGTGCTAGTGTGGTTTTCCCTTTTCGCCGTGGAAAATATGAAAACCTGA
ATGAAAAACTGAATCGCAGTCAGCCAGAGCCGAATTGGAAAAGAGTAACTCG
CATTGGGGACACGAAGAGGTGTCTCGAAAAAGGTAAAATCTTTACACAGAA
ACGACGCCAGAAAGCGATTAGCGATTTNTGACTATGTGTGAGTGTGTAATTTT
35 GGTCTACGGCTGTGTGTCTGCATTTTATTTAACNTTTTGTTCCTCCTGCTGCTC
CACNGTAAAAATAGCTAAAAAAAAGGGCAAGTACTCTTGGCGCGCTCTCCC
TCTCTCTTTGTTGGTCGTGACTGCGACGTCACCGTTACGTTAGTAATCGTTTTCA
AGTGGCGTTTCTTTCTTTCTTTTAATGTGCTGCTTCTTGCTTCTGCCTCTTCTTC
TTGCCTTTGGCTATCTGCTTTGTTTTGAAATACGTCCATGTTATCCAGTGTCTG
40 TGCCAAATGTGTGCGANATGATCTCTACTT

Genomic hit, Accession No. AC009732

Associated ORF

45 Genscan ORF1 predicted sequence
>/tmp/aaaaafra|GENSCAN_predicted_peptide_2|456_aa

MQTKGPITDADCIRGMACRALAGLARS DRVVRQIVSKLPLFASGQLQTLMRDPILQ
 EKRAEHVIFQKYALELLERVSGKTKPLNNPLDPSLSNMHKA NVIAQTRIQYNKQQ
 LYQLIFEHLESNGLSQT AQMLQREVGLPLQTPTRSFHQSPFDYKSLPSGSSSLSRN
 RLRSRMQDVNA AIMGNGDLNRSFGEDSSPAGAGGSNAGDGV SIPNFSSLNTTQTP
 5 IKIRRTDRSSVSRSIQKQAMEPGGMSVGLAEDGQLHPKRITLNTIVTEYLTNQHSL
 CNNPVTTCPQFDLYEPHKCPDPKPSRLSSNYNLTSRHARTQAGFNTSRFDRRYV
 HTHFSPWRSIRSADYEDLEFTCCDLAKYIIVGTQQGDGRVFNMDGVEQFFSNC
 HNFSVDAIKANRAGDLVITSSFWRTPTSILWSIADDEFKLKLRLPDVTYCEFSQTV
 QDRLLGTQNEVY

10

>/tmp/aaaaafrla|GENSCAN_predicted_CDS_2|1371_bp

atgcagaccaaaggacccattacggatgcggactgtatacgtggaatggcctgtaggccttggcgggactgtctgctccgatc
 gggtaggcagatcgtcagcaagcttcactcttccagcggacaactccagacgctgatgcgggatccatactccaggaga
 agcgcgcggaacatgtaattctccaaaagtagcattggagtgctagaacgagtgctgggtaagacgaaaccgctaaataatcc
 15 ttgatccatcgtctccaaatgcacaaggccaatgtaatgccagacacgcatccagtataacaagcagcagctgtatcagc
 ttatcttcgagcacctggaaagcaacgggtctctccagacagccaaatgctgcaacgggaggtgggtcttccgtacagactcc
 cactacgcgcagtttcatcaatcaccttcgactacaaaagcttccagtggtagtagctcgctgctgaaatcgtctgcaagc
 cgcatgcaagatgtgaacgcagcgataatgggcaatggagacttaaacagaagtttggtaggactcctcgccggcaggagcc
 ggtgtagcaatgcgggagatggagtcagcataccaaatttagctcccttaacacaacgcagacgccataaaaataaggagg
 20 acggatagaagttcagttagccgtctatccagaagcaggcaatggagcctggtagtcagttgggtcttccgaagatggta
 actgcatcccaaggatcacccctaaataaccatcgtaacggaatacctcaccaaccagcactcgctgtgcaataatccgggtgaca
 acctgcccgcagtttgattgtacgagccgcacaagtgctccagatccgaagcccagccgattgtaagctgaactacaacctga
 ctatcgccgatgctgaacccaagccggatttaataaccagtcgctttagccgtcgctatgtgcacacgcactttaccatggcgta
 gcattcgatcgccggactacgaggacctagagttcacctgttgcgatttggcggtaatacatcattgtgggcacgcagcagggg
 25 cgacgggacgagtggtcaacatgaacgatggcgtggagcagttcttccaactgtcacaactttagcgttgatgtattaaaggtaat
 agagccgggagacttggtcatcacatctagcttctggcgacacccaccagcattctatgtgtattgaggacgatgagttcaagta
 aagttgcgacttccgatgtcacgtactgtgagttcagtcacacgggtgcaggatcgttgggtggcaccagcaatgaggtatactaa

corresponds to CG10082

30

Drosophila EST several including SD04293 (AI532704)

Annotated *Drosophila* genome genomic segment

AE003454

35

Annotated *Drosophila* genome Complete gene candidate CG10082 – novel protein with
homology to enhancer Pi
uptake

Human homologue of Complete gene candidate

1665793 dbj|BAA13393|
(D87452) Similar to
S.cerevisiae YD9335.03c
protein (S54640) [Homo
sapiens] (2e-43)

40

45 Putative function Putative phosphatase or enhancer of Pi uptake protein

Confirmation by RNAi Reduced G1 and G2/M peaks indicating fewer cycling cells

Example 57 (Category 5)

Line ID 131/8
Category 2nd chromosome, small imaginal discs
Reversion R
Map Position 60A

Rescue ID BamH1

Rescue Sequence 1

10 CACGATTGCNGGCCCATCGAAGTGTGGGTCTATCGATACTCGTGGGTAAATAA
 ACAAGTTCTGAACTGCGATTTTCGGGGGTTTGAGGGGTCAATTGTCCCCTGTGT
 TGAATGTGTTCTAAATCTACACAAACACTCCCTAAGCTTATCCTAAACTTAT
 AAATATTGGTTGCTATTTAAACCCCATTTACGGTTATCCAGCACGCCCCTGA
 ACTGTGACCCACATCCCCGATTTTAGTGACTAGTTTTATACTTATCGTGGTTGG
 15 CATTTGGTACACTACACTTTCTTATTACCTAGATCGCCGACTCCGCGCACGGT
 CGCGCTCCCGTTCCCGCTCCCGATCTCGGCTGCGACTGCGGTGCGGATCCCGTT
 CCCGGTCGCGGCGACCGGCGCCTCCANATCCGGATCCCTAANCGGCANCNGT
 CNTGGTGGCAATCNNGGAATGTTCCGGGGNCCNCTACCNCAGTGNAATCAC
 TGGTACGTCCCACCGCNAAACTCCGCCCANTGCGGTTGCCGGAACGGGTGGC
 20 ANTGCCAATGGGTGCTGCAGAAGGTACCATCACAGCAATCGCTCACGGANC
 CCGAAGACTGCCTCTGCCGCCCCGGTGGGCCACTCATACAGCTACACGGTCG
 GAAATACTACATTGATCACAATGCGCATACCACGCACTGGAATCATCCGTTGG
 GAACGC

25 **Rescue ID** EcoR1

Rescue Sequence 2

AATTGATTTCCGGACATATAAACAGAATCCAGAACTCATCCGGCAGCAGGCTC
 AGTCAGGCCAGTAAATCCGAAAAGAGAGTAACCAGCAGGAAAAGAGAATCC
 ACGTAAATACAGAGAAAATGGCTCTACGCGTCCAATTCGAGAACAACGACGA
 30 CATCGGCGTATTCATAAATAACCAACACATACTGCCTGGTGGCCATCGGTG
 GATCCGAGACCTTCTACAGCGCCTTCGAGGCGGAGCTGGGCGACACCATCCCC
 GTGGTGATGCGAATGTGGGCGGCTGCCGGATCATCGGCCGCCTCACCGTGGG
 CAACCGCAACGGCCTGCTGGTGCCCAACTCCACCACCGACGAAGAGCTGCAA
 CACCTGCGTTACANCCTGCCANAACCCCGAAANATTTATCGTGTGGAAGAAC
 35 GCCTGTCCGCGCTGGGCAACGTTATCGCCTGCAATGATTATGTGGCCCTGGTG
 CACCCGGATCTGGACAAGGAGACCGAGGAGATCATCGCGGACGTGCTCAAAG
 TANANGTCTTCCGCCAGACCATTGCCGACAACTCACTGGTGGGCTCTTACGCC
 GTGCTGAGCAACCAGGGGGGCATGGTGCATCCCAAGACNAGCATTACAGGAAC
 AGGACAACCTGTCGTCCCTGCTGCAGGTTCC

40

Genomic hit, Accession No. CSC:AC020517

Associated ORF

45 Genscan ORF1 predicted sequences >22:13:05|GENSCAN_predicted_peptide_4|357_aa
 MALRVQFENDDIGVFTKLNTYCLVAIGGSETFYSAFEAELGDTIPVVHANVGG
 CRIIGRLTVGNRNGLLVPNSTTDEELQHLRNSLPDAVKIYRVEERLSALGNVIACN

DYVALVHPDLDKETEEIADVLKVEVFRQTIADNSLVGSYAVLSNQGGMVHPKTS
IQDQDELSSLLQVPLVAGTVNRGSEVLAAGMVVNDWLSFVGMNTTATEISVIESV
FKLNQAQPATVTTKLRAALIEDISRSRVAGGGGGGGGGSSGGNSSSGPSTSRRTT
5 RNNAAATAADRPKINEADLEGKSPEEVEMLKTMGFCTFDTTKNRKVEGNDVGEV
HVLKRRKYRQYMNRRKGGFNRPLDFVA

>22:13:05|GENSCAN_predicted_CDS_4|1074_bp
atggctctacgctccaattcgagaacaacgacgacatcggcgctcttactaaactaaccaacacatactgcctggggccatcgg
tgatccgagacattctacagcgcttcgaggcggagctggggcgacaccatcccgggtgcatgcgaatgtggcgggctgcc
10 ggatcatcggccgcctcaccgtgggcaaccgcaaccggcctgctgggtgcccaactccaccaccgacgaggagctgcaaacact
gcgtaacagcctgccagacgccgtgaagattatcgtgtggaggagcgctgtccgcgctgggcaacgttatcgctgcaatgat
tatgtggccctgtgtgacccggatctggacaaggagaccgaggagatcatcgggacgtgctcaagtagaggtcttcggccag
accattgccgacaactcactgtgggtgctcttacgccgtgctgagcaaccaggcgggcatgtgcatccaagacgagcattcag
gaccaggacgaactgtcgtccctgctgcaggttccctcgtggccggaacagtgaaccggggcagcgaagtactgccgccg
15 gcatggtcgtcaacgactggctctctcgtgggcatgaaccaccggccacagagatctccgtgacgagagcgtcttcaagctt
aaccaggcacagcccgccacagtgacgaccaagctgcgtgcggccctcatcgaggacatatcgcggtcgagggtcgccgga
ggaggaggaggaggaggcggcgccggaagcagcggcggaacagcagctccggaccatcgacgtcgcgaaggacgacg
aggaacaatgcggcggccacagctgccgaccggcccaagatcaacgagcggacctggagggtaaatcggcgaagaggt
cgagatgctgaagacaatgggattctgcacgttcgacaccaccaagaacaggaaggctgaggggcaacgatgtcggagaaggtc
20 atgtaatcctcaagcgaagtaccgccagtacatgaatcgaagggtggcttcaaccggcgctcgatttcgtggcatag

Drosophila Gene Hit rescue sequence and TBLASTN with ORF1: b(2)gcn
(EUKARYOTIC TRANSLATION INITIATION FACTOR 6
)((X97641)
25 **Human Homologue** BLASTX with X97641: integrin beta 4 binding protein (HUMAN
EUKARYOTIC TRANSLATION INITIATION FACTOR 6)
(NP_002203.1)
Drosophila EST GH08760 (AI109537 similar by BLASTN to X97641
"D.melanogaster b(2)gcn gene.")
30 **Annotated Drosophila genome genomic segment** AE003462
Annotated Drosophila genome Complete gene candidate CG17611 – bcgn benign
gonadal neoplasia homology
to Eif6 translation factor
35 **Human homologue of Complete gene candidate** 6016331 EUKARYOTIC
TRANSLATION
INITIATION FACTOR 6
(EIF-6)(aa) and 4504771
40 [ref]NP_002203.1[pITGB4BP]
integrin beta 4 binding
protein(aa)
45 **Putative function** eukaryotic translation initiation factor 6 (eif-6)(aa)
Confirmation by RNAi Slightly reduced G1 and increased G2/M indicating block in
G2/M

Example 58 (Category 5)

Line ID 135/25
Category 2nd chromosome, small imaginal discs
Reversion NR
Map Position 24A

Rescue ID EcoR1
Rescue Sequence

10 ATAACATGGGCNCTGGTTTTTAAGTNAAGCTCTANTNATTGGCCCCCATTCTTA
 NNCTCTCTCGCTCTCTTCTCGCTCTTTCGCCTGCTCTCTCGCCTGATTATTCTGC
 TTGGTCGGCTGATGGTTTTINGTTTTNATCTGGTGTATTTTCTGCGTAGTTTATG
 ACAAACCGGCTGGTTCCTTGTGTTATTGCCGTATTCTAATATATTCCCCTATTG
 TTCTTATTTTTGTTGCAGCCTGCACACCTCGGAGGTTCTAGATGATAAGGGGTG
 15 TAGCGATGGTGGGGGGCTGTCTTGANGGGCTTCTCGCCTTGAGCTCTTGTATTAT
 CTTTGGTCATTTGTTATTGTTTAATGCACGGCAATATTATTGGTAAACAAGTTA
 GCCAACAGCACTAAACGCCAATCGCATTCTTTTCTAAAAACCAAGTCTATTGT
 CGATCTTGCTAGGGAAATGATGATGACTCAGGTGCAATTGGGATCTTATCTAT
 GGCTGTCTGGGAATCAAGAAGTGTTCCCGCAGAATTCGTGAANTACTGCCGCT
 20 CTCTCCATGGGGCCATTATTTGCACTCGTTTTNCGCGAAATACCATNAATTAGC
 ATAAAGACACGTCGCCGGCAATCGTGACGTAGGCTATNAATGCCTTCTATGCA
 TGTGCNAACTCGCGGAAGCATAGCAATTTGAAGGAACAATATTTTCANTGCAG
 GTTTTAATGGGCTAAAAAA

25 **Genomic hit, Accession No.** CSC:AC014199

Associated ORF
 Genscan ORF1 predicted sequences >20:54:54|GENSCAN_predicted_peptide_3|117_aa
 MSASPTARQAITQVMPMITRKVVISDPIQMPEVYSSTPGGTLYSTTPGGTKLIYER
 30 AFMKNLRGSPLSQTPPSNVPSCLLRGTPRTPFRKCVVPVTELIKQTKSLKIEDQEQL
 QLDL

>20:54:54|GENSCAN_predicted_CDS_3|354_bp
 atgtcgcgtcaccaccgcccgtcaagccatcaccagggttatgccatgatcaccaggaagggtgtcatctcggatccgatcca
 35 gatgcccagggtgtactctcgacgcccggcggaaccctctactccaccactcctggaggcaccacaaactatctacgagcgggc
 ttcatgaagaatctccgtggctccccattgagccaaactccgctccaacgtgccagttgctgctgaggggaactccgcgta
 ctccctccgcaagtgcgtgcccgtcccccacggaactgatcaagcagaccaagtcgctgaagattgaggaccaggaacagttcc
 aactggatctgtag

40 **Drosophila Gene Hit** TBLASTN with ORF1: BcDNA.HL08053 mRNA (AF132557)
Human Homologue TBLASTN with ORF1 and BLASTX with AF132557: eukaryotic
 translation initiation factor 4E binding protein 2 (EIF4EBP2)
 (L36056)

45 **Annotated Drosophila genome genomic segment** AE003579

Annotated *Drosophila* genome Complete gene candidate CG8846 - phas1 translation
initiation factor 4E binding
protein 2

Human homologue of Complete gene candidate CG8846 - 4758260
ref[NP_004087.1|pEIF4EBP2|
eukaryotic translation
initiation factor 4E binding
protein 2 (4e-16)

Putative function EIF4E translation factor binding protein

Confirmation by RNAi Slight reduction in G1 and G2/M indicating fewer cycling
cells

Example 59 (Category 5)

Line ID 141/12
Category 2nd chromosome, small imaginal discs
Reversion R
Map Position 21A/B

Rescue ID BamHI

Rescue Sequence

10 GGCTCTTTTCCAAANAGGCAGTTTCTTGNCCCATTTCTTGGATTGCTTTGTAGT
 GAACTNAATCGTTTTGTTGGTTCCTCTGTCTCCAGTCTTGTGAAAAATTCGTG
 ATAATAATGCCTGGATAAATANTTAAGCATTGGAACGCGGGGAAAAAGGG
 CTAAGTTGTGTGAAGGAAACAATTGAAGTGACCCTTTGTNTATAAACATTCCA
 CGACGTGTTTCGAAAACAAACAAAGATATGCGGAAACAAAGTGTTAATAAAAA
 15 GAGCNAAAAATAGAGAGAGAGTGTGCGGATAAGCGGTTGAGCGAGATAGAG
 AAAATTGTTGATTAAATGTGTGTCNAAATAAAACATCAAGCCGCTTGAACGA
 ACAGTCAGTTAGTTGCTTCTGATAATAACCATGGGAAGCGGCNCGTGTGCTTC
 GCTCCTCGTTACTTATAAAATATTTAAACGTTTGCATTCTTCNTATTTCCGAAT
 TTTTGCNCCCCTGAANCAACTTNGTTAAACTGCAAATAGCAATGCAAACAAAC
 20 GAATAGAAACTGAAATCGACAACNACATGTGAAATTCACAAATCAAATCGCA
 ATTGTCATCCCAAAGATATAGAACAAGCTATAGGGAAGATANAGAATGTAAG
 TGCCAAACTAAAATAAAACAAACAAGAATAACATTTCCACAGGTGTTTTGCATT
 TCAATGCATATTTCCGTGGCGGNTACAAATCTTTTCAAACCG

25 **Genomic hit, Accession No.** CSC:AC017815

Associated ORF

Genscan ORF1 Predicted sequences >17:48:30|GENSCAN_predicted_peptide_2|554_aa
 MSNKKMFNRTTSVSPGQLHYYHTDFYYSMPDLHKTRKMHGVKRVLVFCLMIVIL
 30 PAILIIMPLHLRKTTFADVTPMAESDIEIRAGISSIFCSKHTLRMNSNFNAFQLRNK
 PEIATNRKHRLKKSMTLPDDTLEYWGFLLKGAKVRVKFCSRVDGSRILIIHGR
 ELNLCGLTDHNKNKLGANYAKGHEQVQVFFEDNVEITEEKGNQDVLMEHENHG
 GEDLTEDIPQPQVNPVKQNNSIQPKLIRKKLKGTHHGEHDMHAITDLQGSHT
 EHILNHHDHSSNSPAHHHNSTAHHREHSSNITNEETSРНHIRNEDEDPDQNSSKTH
 35 YSAESPPIRERLKRHNRAHRNQKRODLYDTLYKRSKRENVYDRKTIHGGNAIN
 FTETDESNSVSSFETGLFQCFNGMILLQEFFRPKNECSNPHIMDTSPNKSSMVVHN
 VIEDGYYYYIFYSNDNHVQNEIHAIFDIYKPTYQYSNMSESQSCLNTTNCTFNISFL
 SDEIVVEVPTRDIEHEEDDITNLSTCHPRSEIYAIFPITVLVLILCCSFL

40 >17:48:30|GENSCAN_predicted_CDS_2|1665_bp

atgtccacaaaaagatgttcaacaggactacgtcagtaagtcctggacagttgcattattatcacacggatttctattactcaatgcc
 ggattgcataaaacccgcaaatgcacggcgtgaaaagggtgctggtttctgcctgatgattgtgatactgccggccattcttatc
 attatgccgctgcatgttcgaaagacggtgttgcgacgtcatctatccatggcggagtcggatcattgagattcgggcagga
 atctctgcatcttttgcgaaacacacactgcgtatgaactccaattcaacgctttcaactacgtaataagccggaattgcgac
 45 gaatcgcaagcacattaggctgaagaagtcgatgacattgccggatgatacgtgaatactggggcttcttctgctgaaagggtgc
 caagggtcgagtgaaattctgtcccgctacgatggatcccgcatcctgatcatccatggtcacaggagcttaattcttgcggtct

gaccgatcacaataagaataagttggcgccaattatgccaaaggtcacgaacaggtgcaggtgttcttgaagacaatgtggag
 atcacggaagagaagggaaccaggatgtgctaattggagcacgagaaccacggcggagaggatttgactgaggatattccaca
 gccgcaggtgaacatactgtcaagcaaaacaattctatacagcctaagttaattaggaaaaaaactgaaaaagggcacaattcatc
 atggcgaaacatgatatgcatgtataacagatttgaaggatcacaccatacgggaacacataattgaatcaccatgatcacagctcta
 5 attctccagcacatcatcacaatagtactgcccatcatcgggagcacagttcgaatatcacaacgaagaaactagtcgtaatcaca
 tacgaaatgaagatgaagatccagatcagaattcaagtaagacccattatagtcgggaaagtcgcctcaccgggaacgtctcaa
 aagacacaatagggtagcccataggaatcagaagagacaggatctttacgatacgtttataaaagatcaagaggggagaatgtc
 tacgatagaagacgatccatggaggaaatgctataaattttacggaaacggacgagtcgaattcgggtgccagctttgagacagg
 actatttcagtgtttcaatggaatgatcctgtcgcaggagtcttcaggccaaaaaatgaatgctcaaatccgcacataatggacatt
 10 cgcccaacaagagttccatgggtggtgcacaacgtcatcgaggatgggtactactattatattctacagcgacaatgatcacgttc
 aaaacgagatccacgccatattcgatattacaagccgacgtatcagtactcaaactagagcgagtcacaaagctgtctgaatacc
 acaaattgcacattcaacatcagtttcctttcggatgagattgtggtggtggaggttccaacacgggatggatcgagcacgagga
 ggacgatataaccaatctgatctccacctgtcatccgcgcagcgagatatacgccatctttccattacgggtgctggtgctgatcctt
 gctgtccttctctgtag
 15

corresponds to CG9524

20	Annotated <i>Drosophila</i> genome genomic segment AE003623 Annotated <i>Drosophila</i> genome Complete gene candidate CG9524 - novel His-rich protein
	Human homologue of Complete gene candidate none
	Putative function No homologies which indicate function
25	Confirmation by RNAi Reduced G1 and G2/M peaks indicating fewer cycling cells

Example 60 (Category 5)

	Line ID	146/2
	Category	2nd chromosome, small imaginal discs
5	Reversion	NR
	Map Position	26B
	Rescue ID	EcoR1
	Rescue Sequence	
10	TTTNATCCAAACTGAGANACTNTTGGCCCCAAACTGAAAACCTCGGACTCGGG	
	CGCGTAAGGGAGTCGGTCNTCGGGAGTCGGTCGTCTTTTGTGATCTTGAGAC	
	TGAAATTCCAATTGTTGATTTATCTCTCGGCTGCTGCGCCGCGGCTGCGCTGCT	
	GCAGCGCAGTCCCACTCCGATTTGACCAGCGACCAAGTTTATAAACTTTGAG	
	CCAAAATGCAGCGGCGCACAGTTGTTACCAAAACGTTGCACGCGTCGTGGCCC	
15	TCATCAAAACAAAAAAAAAATATAAGCGAAAATGAAAACGAAATTCGGTTA	
	ACGTCAACAGAAGCTGACAAAAGGCAGAAAAGACCGAAACAAGTTGCAGGG	
	CCAGAGTAAGCCAAGTTAAATGCGAAAGAGAAGCAAGAGNCAAGAAGAAAN	
	AATGGGCACTACATACATATATTATAGCCAGCTAATCTGTTGTGCAGTGCGTT	
	TTATCAGCCNCGAAAAGAAAACGAAAACGAAAAGTCGGTCCAAGTTCGGAC	
20	TCAAAATCCAAACAGAAGAGACTCCATNCCATCAGAGACACGCGGATCTCAT	
	CTCGGTAATGTCTCAATAAAAGTAATCTTAACTGCCGCCGGAATGTTGGAAA	
	AAGTGAATAATTGAAGCGCTTAACGTGTTTCGAAATACGATACATGAGAAGTCC	
	CAAAAAAAAAA	
25	Genomic hit, Accession No. CSC:AC019865	
	<i>Drosophila</i> EST	GH19286 (AI388389)
	Annotated <i>Drosophila</i> genome genomic segment	AE003481
	Annotated <i>Drosophila</i> genome Complete gene candidate	CG11353 - novel with weak
30		homology to sugar acetylase?
		CG7525 - tie receptor protein
		tyrosine kinase.
	Human homologue of Complete gene candidate	CG7525- 4e-23 4557869
35		ref NP_000450.1 pTEK TEK tyrosine
		kinase, endothelial
		>gi 464868 sp Q02763 TIE2_
	Putative function	Sugar acetylase and receptor tyrosine kinase
40	Confirmation by RNAi	Both gave a reduction in G1 and increase in G2/M peaks
		indicating arrest in G2/M

Example 61 (Category 5)

Line ID 155/13
Category 2nd chromosome, small imaginal discs
5 Reversion R
Map Position 21B

Rescue ID BamH1

Rescue Sequence 1

10 GNTTTAGTCCNCTTTTGANAGGGNCTTGGNGNCTTAAANAANNAAAAAAGGG
GNCCCGGCNCCCAGCAAANAGNNTAAACTTGAATGGTTTAATTCGAAAATC
TTTLAGAAATGTCGCCTAATACCTTATCGGTATAGAGTTCACCTCGTCTCCTAA
TCCATATTTTAAGATATCAATATCTATTAACAATTTTATCGTATGATTAGAAA
TTCGCATTGTTTTATTATTCGACCTTTGGGCTTTACATCGACAGCTACTCTCTA
15 TCCAGACAGGAGACTGGGAGAGAGAGCACGATGCTGTCTGAAAGCATGAATG
ATGGATGCTGTGCCTATGTGCGATATGCACGTTGCCTGAGCTAAAACGAAACG
AGATTATTAATCTATCCGCAAGATTGAGATGCTGATTCCACATGAGTGAGCGA
GTCCGTGAGTGGATATTGCTCTCTCCGAAATGCATGCATGAGTGAGCAGGGGG
GCTTCAATCGCNCTNTCGATNTGCGACAGNGACATNTTTTATCTTCGACNAT
20 GCNCTCNCTCCCTCCCACAGAAATCTTGCGCTNGNTCTCCGANNTNGGGNTNG
ANGGCNCTCTTCTCTNTCCTTAAATTGGGANTTNNCTTTTTTCNAANAAGGGN
NAGA

Rescue ID EcoR1

Rescue Sequence 2

25 AATCNTTTTNTCCATTNGGCGNCTTNCTCAAAACATATTCACATTTGGNCCCAA
CGGCGTANGACTTNATCTCACGATTGTTTGGTTTCCTACTCTCCCGCGCTCCCT
CTCTTCTGAGTCTCTTTCTGGCTGATTGCGATTTCGATTTTAGCCGCTGCCATCG
CCGTTGTTTTGCCTACCTATGTGTGTGTGTGAGGAGTGTGTCTTGATTTCAGT
30 CCGCAATGCGCTCCGCTCATTATTTGTTTGANCGCCGCGGTGTAAAGTTGTAA
AAAGTCCAAGTGCTCGTGGAAGTTCGATGCAAGACGGGGAAAACGAAACGCG
ATAAATCGTGAGAAAAGAGAGTGCCTAAAGGAAGAGGGAGTGATAATCAN
ACGAAATGGAATAATGTNTTGCAGAGGCNACAACAACAATGCAAAATAGTTG
TCATTGAGGCGCAATGAATGATAATTAGTGCTTANTTGAAATCATAATCNTGA
35 AGAAAGCGTAAAGCTCGATTNTGGCAATNTATTCTTGATTACCANTGAGTCTG
TGATATTGCCGTGTGTNCCGAAAATGGANGTTATNAAACCCATGGACTTCAGC
ACCTTCTCCGCGTTCTGCGAACATCTTAACAAATCTCCACAAAATTGCAGCAA
CAACTGCANCGACGGTACCGCCAACTATAANCAATGGAAAANGCATTATTTG
GAGGTAANAGCNAAAAATACCAATNTTCCAATGCGAAATTGCNAGCNTGG

40

Genomic hit, Accession No. AC004274

Annotated *Drosophila* genome genomic segment AE003590

Annotated *Drosophila* genome Complete gene candidate CG13693 - novel

45

Human homologue of Complete gene candidate 6e-05 4507659 translocated

promoter region (to activated
MET oncogene)
>gi|1730009|sp|P12270|TPR_
HUMAN POOR MATCH

5

Putative function No homologies to indicate function

Confirmation by RNAi Only wild type profiles observed

Example 62 (Category 5)

Line ID 162/24
Category 2nd chromosome, small imaginal discs
5 **Reversion** R
Map Position 55C

Rescue ID EcoR1
Rescue Sequence 1

10 TTTTNTTTCANGGNTCTTTGCNCATAAAAANACACGNGCCCTCNTGTCCATTTCAC
ATTTTACTTGGAGTCGGTAACGTTGAGTTCGCGTCCGTGCGTTCTGCCTTCCA
ATACAAAGTCTGGTGTGAATCTACCAAGCATTCCAGTGNGAAAATCAACTCAC
ATTGCTCGGTGATCCNTGCGGCGGTATNATCGCACCCGGAATTGCATAAGTTG
CGGNGAGCGGAAAGAGAGTGCACGGATTTCNGTTATCNAAGGGCCGGCANC
15 NGTGGGGCGGCGACGGNAGAGCACGCAGAANAANAATANANTGNNGTGGCG
AATTNAAAAATANNATNAAAGAAAATTCGGGCGGCTAATTTTTCTTCAAATTT
GTGTGCGGTGCGCGAAAAACAACGTGTTTTTCNATGGTTGATAATACACACGG
ACGGNNCACTCGCGCTCACCCACATAGTCACNAAAGTCGGCGACGTCGACGA
CCNCACNCTCATATANGGACNTTTAATCCCGTNCATNCGTGTAGCGTNCNTA
20 TTTAACNTNTCTGTCCATCGGAACGCNCGCNTTCTCGCCTTCNTTCTNCTTTA
CTTTAATTTCTATTTNNAAGGGGNAGNCCNATCTTTTNCCTNTCNNTGCCNT
TTAANNTCATCCACANCTCNCTTTNTCNTTCTCCNCCTTNTNTTCTTTTCNTC
TTNCTTNTGNCCTTGCCTCGTTCTTTCTCTTCNTCTCCTTNCCTTCTCCTCCTTT
TTTCTCCTTCCCCC

25 **Rescue ID** BamH1
Rescue Sequence 2

AAGNCNCCTTGGCCGNNTTNAACGGNAANTAANCCGGGNCCNCGGGNCNCGA
TAATCAGGTCNANCCTTGTGCCTACCACCACCAAATTGAAAAAGAGCNAAGA
30 TTCTCTAAGGCAAAAACTCCCAATCTGTGGAATTTCCGGAAGCGAGAGCAC
ATTCAAAGCTACCAGTTATCAGCGAGCAGCATGTCTAAGCTCAGGAACCTGTT
GCCCACAATCTTTGGCGGGAAGGAGGCACAGAATCCGACACCCGTCGAGGGA
CGCCTGGAATAGGACGCAGCTCCCGTGGACGACAACGAACCNGATTACTACT
ACTGCGGAGCCATGGCGCTGCCCTCCACCGCTGGCACGCCCACAGCCTCCTCG
35 GATCTGACCGAATCCGTGCTGCGCGAGCTCAGCGACCCAAACTACAATTCAAT
GGATGTGGTGTCTTCNNCTNTTTTCCGGGCACTCTCAGTAACGTCCAGACAA
ACAACACCATGAACGTTACNGCGCCAGCAACAGGTGGTCATGAACTTCTCG
AATGCCAATAATCTGCACTTCGGCTCCGTCTTCAACTTCAACCAAAACTTGAG
CGCCTGCNGCTCNCGAANGGGTTTACCNGTTCGCANAAGAATCGGTGCGCTC
40 TCCANACNGT

Genomic hit, Accession No. CSC:AC012981

Associated ORF

45 Genscan ORFs: ORF2 predicted sequences
>18:26:17|GENSCAN_predicted_peptide_7|1320_aa

MEETNNATTIEQQPIALINGQEQVANEQQPSSPTSVA TPTSTTSGGTGNATPAFSY
 DDLFPALPANTS AQSGASGSLARVTSSQKTHIVHVPCKERKSTESEKFGESES
 KRICQQITKETGAQIELASRQVTVPREHFRVILGKGGQRLREIERVTATRINIPSQSD
 ESEFITIAGTKEGIAQAEQEIQLSAEQYKKSSDRITVPKVYHPFIVGPYSENLNKLQ
 5 EETGARINVPPQQVQKDEIVISGEKDAVAAAKAKVEAIYKDMEEKCSTVSVEVAK
 PKHRYVIGPKGSTIAEILQLTGVS VEMPPNDSPSETITLRGPQVALGNALTVVYQK
 SNSVKSVEINAAHWHKYVFRKGANMKQLEEDCPNVNVNCLDKIKLEGDPEN
 VDRAYAYLSEIKNYEENFTFEVMTVNP SYKHIIGKAGANVNRKDELKVNINIE
 EREGQNNIRIEGPKEGVRQAQLELQEKIDKLENEKSKDVIDRRLHRSIIGAKGEKI
 10 REVKDRYRQVTTITPTQENTDIVKL RGPKEVDKCHKDLLKL VKEIQESSHIEVPI
 FKQFHKFVIGKGGANIKKIRDETQTKIDLP AEGDTNEVIVTTGKKENVLEAKERIQK
 IQNELSDIVTEEVQIPPKYYNSIIGTG GKLISIMEECGGVSIKFPNSDSKSDKV TIRG
 PKDDVEKAKVQLELANERQLASFTA EVRAKQHHKFLIGKNGASIRKIRDATGA
 RIIFPSNEDTDKEVITIGKEESVKKAREQLEAIKECDEVTEGEVSVDPKHHKH FVA
 15 KRGFILHRISEECGGVMISFPRV GINSKVTIKGAKDCIEAARQRIEEIVADLEAQT
 IEVVIPQRHHRITIMGARGFKVQQVTFEFDVQIKFPDRDATEPVEGLTNGGSGENG
 GENEGQEGEQEVEKEAEQEPVRQCDVIRITGRIEKCEAAKQALLDLPIEEELSVPF
 DLHRTIIGPRGANVRQFMSKHDVHVELPPSELKSDVIKVCGTPARVAEAREALVK
 MIEDYEADRADRELRSFVLQVDVDTEFHSKLIGRHGAVINKLRADHDVIISLPKRD
 20 EPNDRIISITGYQANAEAARDAILEIVGDPETLHREVIEIDKRIHPHLIGQRRRTIRKII
 EDNKVNIKFSADDDNPNISIFISGKIEDVENVKELLFGMAEDYERDYLDNVAIAPPTI
 GAFLTGFWIRCRRCQRRIRHQRRTVGEAKAGQKPDCAQHSVAGGLPALRCWRG
 SGGHLHAYHLRVGPQKLSASGRVSRSPAVAAILQVGVRRGSELEMDQELEQKLELE
 LELDYRAMSGRAAAVVRTSL

25

>18:26:17|GENSCAN_predicted_CDS_7|3963_bp

atggaggaaactacaacgcaactaccatcagcagcagcccatcgctctcattaatggccaagagcaggtggccaacgagca
 gcaaccatcctcgccaactcagtgggccacgcccactagaccactagcggcggaactggcaatgccacacccgcccttagctac
 gacgacctgtttccggccctgcccggccaacactcggctcaatcgcaatcgaggcttcggctcgactctagctcgtgtgacgag
 30 ttccaaaaaactcatattgtgcatgttccctgcaaggagcgcaagtcacggagtcggagaagtttggcgaaggcgagtcgaag
 cgtatttgcagcagatcaccaaggagaccggagcccagatcgagattgccagtcggcaggtgaccgttctcgggagcacttc
 cgcgctatcctcggcgaagggtggccaacggctgcgcgaatcgagcgtgttactgcgacgcgcatcaacatccccagccagag
 cgatgagagcgagtttatcacgattgccggaaccaaggagggtattgccaggccgagcagggagatccgtcagctgtcagccg
 agcagtagaagaagtcacggaccgcatcacgggtgccaaagtttaccatcccttcacgtgggcccctacagcgagaacctaaa
 35 taagctgcaggagagaccggcgctaggatcaacgtgcgcgcgagcaggttcagaaggacgagatcgtcatctcgggagag
 aaggacgcggctgcagcggcaaggccaagggtggaggccatttacaaggatatgaaaagaagtgtctaccgtcagtggtga
 ggtagctaagcccaagcaccgatatgtcattgtccgaagggtccaccatcgccgagattctgagttgaccgggtgtgtctgtag
 agatgcctcccaatgactccccctggagacgatcatttgcgtggccgcaagtggtttggaaatgcctaaccgtgtgtctac
 caaaagtcacactcgtcaagtctgtggagatcaatcgccacattggatccacaagtatgtgttcggtcgaagggggccaaca
 40 tgaagcagctggaggaggactgccccaacgtgaacgtgaattgcctggaagacaagatcaagctggaggagatcccgagaa
 cgttgacaggggtgtagcctactgtccgaaatcatcaaaactacgaggagaactcacattcgagggtgatcaggttaaccttc
 gtactacaagcacatcatcggttaaggctggagccaacgtaaatcgctgaaggatgaactgaaggttaacattaacatcgaagag
 cgcgaggggccagaacaacatccgtatcaggggtcccaaggaggagtagcggcagggcgagctgaattacaagaaaaaatcg
 acaactggaaaacgaaaaatgaaggatgtgatcagaccgcccgtctccatcgttctattatcgagctaaggggcgagaagatt
 45 cgcgagggtgaaggaccgctaccgccaggttacaatcacgatactacgcccaggagaataccgatattgtgaagctgcgcgg
 acccaaggaggatgtggacaagtgtcacaagatctgcttaagctggtaaggagattcaggaatcgtgcacattatcaggtg
 cccatcttaagcagttccacaagttcgttattggcaaggggcggttaacatcaaaaagatccgcatgagaccagactaaaat
 tgatctgcctgcgagggtgacaccaacgaagtgtatgaatcaccggcaagaaggagaacgtgctcaggcggaaggaaacgta

tccaaaagattcaaacgagctttccgacattgtcaccgaggaggtgcaaatcccgcccaagtactacaactcaatcatcggcact
 ggccggcaaacatcatctcctcgatcatggaggaatgcgggtgtgttctatcaagttcccaacagcgactccaagagcgataaggt
 cactattcgcgggtcccaaggacgatgtggagaaggctaaaggtcagctattggagctggccaacgaacggcagctggcttctt
 accgccgaggtgcgcgccaagcagcaacaccacaagttcctgatcggcaagaatggcgcttctatccgtaagattcgcgatgcc
 5 actggtgcccgcattatcttccctcaaacgaggacactgacaaggaagtgtaccatcattggcaaggaagaaagcgtaaaga
 agggcccgtagcagctggaggcgatcatcaaggagtgcgacgaagtaaccgaaggtgaggtttctgtcgtccaagcaccac
 aagcacttcgtggccaagcggtgcttcctcctgaccgcatttcggaggagtgccggcggtgatgatctctcccccggtcgg
 catcaactccgataaggtgacgatcaagggtgccaaggactgcattgaagcgcccgccagcgcatcgaggagatcgtcgccg
 atctggaagcgcagaccaccatcgaggtggtgattccacagcgctcatcatcgaccatcatggcgccacgtggttaaggttca
 10 acaagtacctttgagttcgatgtgcagatcaagttccctgatcgtgatccaccgaaccggtcgagggttgaccaacggaggc
 agcggagagaatggaggcgagaatgaaggccaggaggagagcagggaagtagagaaggaagcgaacaggagcggttc
 gtcagtcgatgttatccgaatcacgggcagaattgagaagtgagggccgcaaacaggctctgcttgatcttccccatcgag
 gaggagttgctggtgcctttcgaactccatcgatccatcatcgaccgcgcggtgccaatgtgcgtcagtttatgtccaagcagat
 gtgcacgtagagctgccacctagtgcagcttaagtcggatgtgatcaaggtctgcggtacgcccgtcgcgtcgccgagggccgc
 15 gaagcgcgtggtgaaaatgattgaggattacgaggtgataggccgatcgtgagctgcgtcctttgttctccaggtggacgtaga
 tacggaattccattcgaagctcattggtcgtcatggcgctgtgattaacaagctgcgtgccgatcacgacgtcatcttcgtgcct
 aagcgggatgaaccgaatgaccgcacatctctatcacccggctaccaggccaatcgaggagcagcccgcatgccatcctaga
 gattgttggcgacccccgagacacttcacgcgaggttatcgagatcgataaacgcaccccccacctcattggccaacgcgga
 cgcaacttcgaagatcatcgaggataataaggtgaacatcaagttctcagctgatgatgacaacccaattcgtatctcatcagt
 20 ggcaagatagaggacgttgagaacgtcaaggagttgctcttcggcatggctgaggactacgagcgtgactacttgataacgtg
 gcgatagcggcgccaacgattggtgccttcctaactgggttctggatccgatccgaggtgccagcgagaacggttcgtcatc
 aaagacgcaccgtgggagaagcaaaagcaggccaaaacctgactgcgccaacactcagtcgaggaggacttcccgact
 tcgctgctggcggggctccggtggcctccacgcctatcacctccgtgtggggcccaaaaactaagtcacgggcccaggtgtc
 ccgatcggcagcagtagcagcaataactacaagtcggggtgcgcggggatcggagctggagatggaccaggagctggagca
 25 gaagctggaactggaactgaattggattatcgggcaatgagcggcagagcagcggcagctgctcgacatctcttag

Drosophila Gene Hit BLASTN with rescue sequence 1: dodeca-satellite protein 1 (DDP-
 1) (AJ238847). TBLASTN with ORF2:dodeca-satellite protein 1
 (DDP-1) (AJ238847).

30 **Drosophila EST** GH20785 (AI389573), LP07358 (AI294065)

Annotated Drosophila genome genomic segment AE003799

35 **Annotated Drosophila genome Complete gene candidate** CG5170 - Dpi dodecasatellite
 DNA binding protein
 CG5576 - Bg5 involved in
 cytoskeleton organization and
 biogenesis which is putatively
 a component of the plasma
 membrane

40 **Human homologue of Complete gene candidate** CG5170- 4885409
 ref|NP_005327.1|pHDLBP|
 high density lipoprotein
 binding protein
 45 >gi|2498434|sp|Q00341|HB

CG5576- 2e-07 4506539
ref|NP_003795.1|pRIP|
UNKNOWN >gi|3426027
(U50062) RIP protein kinase
[Homo sapiens]

5

Putative function CG5170: DNA binding protein (homology with Scp160p, a new
yeast protein associated with the nuclear membrane and the
endoplasmic reticulum, is necessary for maintenance of exact
ploidy)
CG5576: death domain containing protein, possibly involved in
signal transduction

15

Confirmation by RNAi CG5170: Reduced G1 and G2/M peaks indicating fewer
cycling cells and more polyploidy
CG5576: Loss of G1 peak

20

Example 63 (Category 5)

Line ID 40/2
Category 2nd chromosome, small imaginal discs
Reversion NR
5 **Map Position** 39B

Rescue ID BamH1

Rescue Sequence 1

TTTTGCCTCCGCTTTTTTAATTAATAAAAAATGTNTGTTTNGCCCTGGAGCTCTCG
10 GTCTGTTAGCGAGCGTTGCCACCTTTCTGCGAGCTGTTGCTGCACACTGCCACT
TTACGAACACAGCTCTGATAGCGGGACAAAATACGTCAAGGCAGCGACGGTG
GGTTACTAGTGAATTTGGAACGGTGGTCTTAAGACGTACTGGTCTTTTATATTT
TCATTATTTTTTAAATTGTCGCTCATTTACCAATAAACCTTTTACTTTTTCCTG
ATAGTCCGAAGTCAGATCAAATAGGAAGTTTCACAAAAAATTTTCATCCAGAG
15 AAAATACGCCGACGCTATTCGAGTTTTTGTATTTCGTTAACCGGGAAAGAATA
GTTCGAATTCGTTTCGCACTTTATCGATAGTAGATTGCTATTATGGAGCCCACTA
GTAAATTAATTAATTCAGACTGATAAAAGCGATCAACTTTTGTTAATGGGT
TTAANTCTATAATAATNCTTAGTCCAAATTGTNTCAAAGTAGTCGATAATTTAT
AATAACAGTTTTAGATGACCTCTAGGAAATAACTAATTACCCACATNCTTCAA
20 GAAAGTGTNTNCAATTTGTNCTATAATTAATAACAGTTGTATTAATTATGTTG
TNATTGTNACTCATAATACAAATTAACAATATAAACACACATAAATAAGAG
AATTGGAATATTTTGTCTCAGATTAGATTTNCCAC

Rescue ID EcoR1

Rescue Sequence 2

AACGGGGGGGCTTCCGCGNCNCCAAAACGCAATNTACCGTTCATGCTGTGAAG
CGAAAAAGAGTGGTAGCGCCTACCNTGGCATATGTAGTTAAATCCGTGAAAT
AAGTGAATAAGAATATATGTATGTACTTAATTCGAAAACCTTTTCGCCGTCAG
CACAACGGGTGAACGAGAGAGCGGAAGTGGAGTTTTTTGTGGCGGGTCGTCT
30 CGCTCGCACCGCAAANGTCGTCCGTGGCTGCGTGTATGGGTGTGTGGAAAAA
GCGTCGAGGTGAATGTGGATTTCTAACCACACCAGCATTGCAAAGACATTGAT
TGATATTTAAAGCTGCAGCAGCGAACAAGCAAATCCTAATTTTCGGCAAAGTT
TAAGAATAACGAGTGACTGGGGCGCGCGCAATAAGATAAAATTGAAGGTTAT
CTGTGTGCGTGTGAGTGACCGTNTACCAGTGTGTGTGTGCGANCGTCCATTGT
35 AAACAAAAACAAGTGTGTGAGCGGAGAGAAGAAAGGGAAAGAGAGAAAG
AGCGAACAGACTGGCGAGAGAAAAAAGAGATGCCACAAANAAAGCAGCGCA
CAAAGGAAAGCTGAAAAATTCANTAAATCTGCAAAAGTGAAGAAAACCACAA
GAACCCGCAGTCNTGTTAAATAAAACCCAGANTCCAAGAAACNTTAAAGAA
GCAGTGCAAAACAACTGGTGCTNTGAATGCGGTTTATTTTGAATAAAATGCA
40 ATTCGGTCCGATGGAA

Genomic hit, Accession No. CSC:AC014744

45 **Drosophila EST** several including LD46342 (AI544109 BLASTN similar to mRNA L07550)

	Annotated <i>Drosophila</i> genome genomic segment	AE003669
	Annotated <i>Drosophila</i> genome Complete gene candidate	CG8678 - novel with ankyrin homology
5	Human homologue of Complete gene candidate	CG8678 -gi7661580 B69CEC399B56F35C ref NP_056425.1 DKFZP434J 154 protein [Homo sapiens] (2.20E-85)
10		
	Putative function	Novel protein with ankyrin domains, unknown function
15	Confirmation by RNAi	Reduced G1 and G2/M indicating fewer cycling cells

Example 64 (Category 5)

Line ID 55/12
Category 2nd chromosome, small imaginal discs
Reversion NR
Map Position 49C

Rescue ID BamH1

Rescue Sequence

10 TCTCATGNTCAGGGGGCCTTTACNATGTCAAAGAGCAAATTGTCCACAGGGCA
 GCAACCGCAAGTGAGAGACGGGTGGAAAACCTGGGCGGCATGACCATGAATGA
 AAGCCGCGACCGGCAAACGTGGCCCGCCACAAAGCGAGCATTTTACATTTT
 AACTGTCTGGACATTTTGAAGTTACACCAAGGCAATGATACCAGTAAAAAAG
 AAGAAACAATCATTTTGAATAGATTAATCACCTGATTAATGTTGGTTGTATGT
 15 TGATTGTAGGTGTTTTAATATACAATGTCTCTATTACTGCTTTCCTTTATTCAAA
 AGCCATGTGTAAGTGTAAGTTCTCGATTTTCGGCTAGATTTTGAAGTTCTGCCAT
 TATCAATTAAAAGTCCAGTTCCTCTATAAATTGGTAATAAAATAGCTCTTTACA
 GCCAAGTATATGTGCAATTTTGTAAAGATTAAANGTCCAAATGTTGTGAACCTT
 TCCTGGCCCTGAATTTTAAAAAACCATTAATTTGGTCCCATTTGACATTAAATG
 20 TTCTATGTACATTAATATGACTTTTTGTGGATGGTTTTATAACAAGCATTACT
 ATATTCTAAAAATCAAGGATAAAGGACNAGCTTTACAGGAGGTAACATTCCTA
 TTGTACGGCTTTATTTTCTTATACCCATAAGAGCATACCACTAGGATCCGTCGA
 CCTGCAGATCTCTAAAACTTGCCTTTGCTGGCGTTTTCCATAA

25 **Genomic hit, Accession No.** AC007085

Associated ORF

Genscan ORF1 predicted sequences >21:54:11|GENSCAN_predicted_peptide_3|108_aa
 MGLVTAAFKLKRKDIQDRYQHDIRICHTRSTAHTAYAHFAEHLRLRSPRQRFVN
 30 GKGAALVLLLVSAARQFSGSTGAYKLGNRVVGKVEGEQQEYKLQDRTHFCGN

>21:54:11|GENSCAN_predicted_CDS_3|327_bp

atggggctggtaaccgcccgttcaagctgaagcgcaaggatatccaggacagatatcagcatgatattaaccgcatctgccaca
 cacgtagcacggcacacacggcgatgctcatttgcggagcatctgttcgacgaagtccacgtcaacggttgcacacggcaa
 35 aggtgctgcgttgctcctcctcgttctgcggctcgacaatttctggctcgacaggtgcctacaaactgggtaataagagttg
 gaaaagtagaaggggaacagcaggaatacaaaactacaagacagacaacacatttctggcaattaa

Corresponds to CG8732

40 **Annotated *Drosophila* genome genomic segment** AE003836
Annotated *Drosophila* genome Complete gene candidate CG8732 - l(2)44Dea
 homology to fatty-acid-
 Coenzyme A ligase, long-
 chain previously described
 45 spindle/chromosome

abnormalities in neuroblast
squashes

5 **Human homologue of Complete gene candidate**

1e-171 4758330
ref|NP_004448.1|pFACL3|
fatty-acid-Coenzyme A ligase,
long-chain 3
>gi|4165018|dbj|BAA371 and
LCFD_HUMAN LONG-
CHAIN-FATTY-ACID--COA
LIGASE 4 1e-157

10

Putative function Fatty acid CoA ligase

15 **Confirmation by RNAi** Only wild type profiles observed

Example 65 (Category 5)

Line ID 6/7
Category 2nd chromosome, small imaginal discs
5 Reversion NR
Map Position 28E

Rescue ID BamH1

Rescue Sequence 1

10 TATNAATAATCATAGGGCTCTTGCTCTTACGTGTAAGGCCTGCCCTCTNCCA
GTCTATATACAAAGAAAAACACACACACACTGGCACACTGGTGTTCGCATATG
CCAAAGCCGAGTTAATTTTCACTTTGTTTAACTATCGTTTGGTGTGTTTTGCATTT
TTTAACCGCGCAAACGGTATTTGCGCGTTTTGCGCCTCTTACTTTGCGATTTAT
TGCACCGCTTGGCTGTGTTTTGCAATTTCTATCTTGATTTTCATTGGTATTCACG
15 CGTAATGTAATTCCTTAGCAGCGTGACCGCGCCGATAACGATAAAAAATACCAC
GGGACCAAAAATAAATACCATATGATACCACTTCAGGGAAAAAGAAATCCTAT
TTAATACCACTCACTTTAAAAATAAGTTTTTAAAAATATATATNTTTATTTAAA
AAAAGGTGTATTTATAATCAAATACTCGGTACTTNTTAATTACTCCAAGAANA
ATTAATTTGAAAAAAAGGGGTTCCATTATAAAATATATATTAACCGCTTACAC
20 ATAATCCCCAAACAAAACAGCGATTGGGATTTAAAAGGTTCTAAGTCCATCAT
TATAAAAGATCATTTCCGAAAAACAAAAGAAATAGATTCAAAATTAGGCGAC
ATCAGCCCGCTGATAANGATCATAAAAATACAGAAGCTNATTACGCGAATCA
GAAANTCCTACTCGCCACTATCCGAAAACNTNGAAAAAAAATGG

25 Rescue ID EcoR1

Rescue Sequence 2

TGAAAGGTAGCAACAACGTTTTCCTTGAAAAAGCTGTAAATAGTAAACAAAA
TTGTCAAGTTAACGAGCCAAAGTTATTAATAAGGTTTCGAGTACGTTGGCATC
GGCTGCCCAGGCAGCAAANAAAAACAAAGACGCAGTTCAAGATCAGCTGGAC
30 ACTTAGAAGANTTTAAGAATTGAAGCACATTNNAAGAAGANAAACAAGAAC
CCCACCAAAAACCCGCGTGCGTTTGTATGTGTGTGTGCCATCAAATTTCCCGC
ACTGGGTGAATGTGCNTGCGTGTGTTNTGTGTGTCATTTAATTTTCCTACCAATAA
TCGCCTTCCAAGAAGTGAATACCAGCCGATCCACCGCTAAATCGAAAAAAGTT
TNACTCTGGGTAAANTCACTGTTTACGGCTTTTGTGCTATAATTACCTTTCCCG
35 TAAGCNGTGGGAANCTAAAANCCAAAACNTNAGAATCCGAATTCCG

Genomic hit, Accession No. CSC:AC017934

Associated ORF

40 Genscan partial ORF1 predicted sequences

>22:35:21|GENSCAN_predicted_peptide_4|128_aa

MGTNSGATAGINNKPVGGATGAGVLVGGGVGGANSSIGGVLSNSLGGGGSGGSL
ISGLNAGGQNANVGGMGNVGGDDGGNGMVGGGVNNQATTTPQYTIPGILHFIQ
HEWSRFELERSQWDVDRAELQ

45

>22:35:21|GENSCAN_predicted_CDS_4|384_bp

atgggcaccaattcgggagccaccgtggcataaacaacaagccggtggcgggtgcaacaggagccggcgtcctttagggc
 gcggtgtggcggtgccattctcgcgcgggtgtcctgtcgaacagcctggcggtggcgccagcgcggtctgagcatc
 agcggcctcaacgctggtggacagaacgccaatgtggcggaatgggcaacgtggcgcgacgacggcggaacgggatg
 gtggcgcggtgtaaataaccagcaggccacaacgccccatacacaataccgggcatcttgcacttcacagcagcagtg
 5 tcgcgttcgagctggagcgatcacagtgggacgtggacagggccgaattgcag

Human Homologue TBLASTN with ORF1: very weak homology with striatin,
 calmodulin-binding protein (STRN) (NM_003162.1)

Drosophila EST several including LD42534 (AI516610), LD03224

Annotated Drosophila genome genomic segment AE003619

Annotated Drosophila genome Complete gene candidate CG7392 – novel WD40 family
 member

15 **Human homologue of Complete gene candidate** CG7392- SG2N_HUMAN
 CELL-CYCLE NUCLEAR
 AUTOANTIGEN SG2NA
 (S/G2... 622 e-178 A cell-
 cycle nuclear autoantigen
 20 containing WD-40 motifs
 expressed mainly in S
 and G2 phase cells

25 **Putative function** WD40 protein a novel nuclear protein mainly expressed in S and
 G2 phase cells that was characterized using autoantibodies from a
 cancer patient

Confirmation by RNAi Reduction of G1peak, more polyploidy

30

Line ID 103/1
Category 2nd chromosome, small imaginal discs
 35 **Reversion** R
Map Position 57B

Rescue ID BamH1

Rescue Sequence 1

40 GATTTCAAAATTAGGCGACATCAGCCCGCTGATAAAGAATCATAAAAAATACT
 GAGGCTTATTTTAGCGAGTCAGAGACTCCTACTCGCCAACTATCGAAAACATA
 GNGAAGATATAGTCGCCAACCGATCTGCCTTCTATAGTGTTGCTTATTGTTGTC
 CCCTAATCAAATTAATAAAAAATCTGCATTAGGCTGCTTCGCCGGCCAGCAACA
 AATGTTTTACACCTACTGTACTTTTCGCAGAACAGAGATCCAAATGCAGGATC
 45 GTTTCATGACTGTACATTTATTCGGATTAGACATTAAATTACACCCTACAGCT
 ATACATACTAACAGTGAACACGGCAAATGCTTAGCTAGCATTGGGCCACTTTC
 GTTGACTGCGAATAAAAAATGATTGGCCGATGCCTTTAGCAGATTCTTTTGAT
 CGAATTACTCGGATGGCTTGTGTGTCCACCTCTTACAAGAACTCCTCGCACCA

ATCGTTGAGACAGTTGTAGCAATCGGATGCTTGGTTGGAGCTGGCGTGGCACA
CCTTCTTCATCCAGTCCTTGGACAGNTTCTTGGNCCTTTTCAGNANCAGGATCT
GGTCCCAAACGGNGGAAGGCCTAAAACGAATGGNAATTGATCGGTAGCCCTT
GACTGGCATTGGTAATTTGCGCACATGGGNGTCATCGGATTTACACACGCACC
5 ATATCGAATCAGCGTCCTTAAGCGTCAACCGAGGGTTTCCCAATTCCGGCCA
GTTCCGTCACCGACTTGTTGCCATTGG

Rescue ID EcoR1**Rescue Sequence 2**

10 ATCAAAGCGNCTGGGCCCCGTGCATCGCCNCAGCGTTCGTCTTAATTAATTAGT
GATTGCAAGCGGGTGCAATTATGCACAAAATTACGGACTAATACAACTGCCC
GCTTCGCGCTCTCTCCATCTCCCTTCCAAATAGTCGTTTGTCTTCGCACACAA
AAGTGTAACCCCTGTGAAAGGTAGCAACAACGTTTCCTTGGAAAAAGCTGTA
AATAGTAAACAAAATTGTCAAGTTAACGAGCCAAAGTTATTAATAAGGTTCTG
15 AGTACGTTGGCATCGGCTGCCAGGCAGCAAAGAAAAACAAAGACGCAGTTC
AAGATTGAGCTGGACACTTAGAAGAGTTTAAGAATTGAAGCACATAAAAAAG
AAGAGAAACAAGAACCCACCAAAAACCCCGCCGTGCGTTTGTATGTGTGTG
TGCCATTCAAATTTCCCTGCACTGGGTGAGTGTGCGTGCGTGTGTGTGTGTGTC
AGTTTAATTTTCCCTACCAATAATCGCCTTTCCAAGACGTGATTACCAGCCGATC
20 CACCGCTTAAAATTGATAAACGTTTTAACTCTTGCGTTACATCAGCTGTTTTAC
GGCTTTTTGTGCTATAAGTTACGCTTTTCCCGTAAGCCGTTGGCAACACTAGAA
CGCAAAGAGCATAAAGAATCGCGAGTACCGTANAGAGGAAGAGAGGAAGA
GAGAGAGATAGAGAGTGTGAGCGTGTGAGTGAAGCGGGGAATGTGGGGGCGGT
TCCGGTGCGAAAAACGTAGTAGTAGTACATNNAGAGAGTGCGAACGAGAGG
25 GAGGCAGCCAGCGAGTGTCTGCGACTGCTCCCCCTTTACCCTCGTCGCTTTT
CTATTGCGAAAAATTCAATGACCTCATTTGTTTCATGTGCCGAACTTTGCTTTTC
TTTCCCAACCTAAAAACGCAAAAAAAAAAAAAACNCCAAACAGGATATACGTNG
GAACANTGANCAAAACNANTTCGANAAAAACCAACAACNANGGACCGTGCCCTG
GGGCNCCTGAAAGGCAAACAGCTGGCANNCAAATCCGGAAAAGGATCNGGAA
30 NAACAGGATCNGCGGGCNCAAGGATCNCCGGAACAGGCAAAGGAAACNCCC
GGCNCACNGCACAAGCCNCTGAAAAGCAACNTGAACCAATGGGCACCANTTC
CGGGANCCACCGCTGGCATTAAA

Genomic hit, Accession No. CSC:AC017934

35

rest of results as for line 6/7

Example 66 (Category 5)

Line ID 65/24
Category 2nd chromosome, small imaginal discs
5 **Reversion** NR
Map Position 48A

Rescue ID BamH1

Rescue Sequence

10 TACGATTTTTGCANTGCNCCATTTCTGTTGGCACCCGATTTGTATATATATTTTTT
ATATAACCCACGGATTGCCAACTTTTCATTGCCCTTTCACACTCTTATTCGCCAT
TTATGAACTCTTCTTTGACGATTGGAACGGTCTTTTTTCGCTATTTTCGACTGC
ACCCGCGCTCTTTTCGCTTCGCTCTCCTCCCTCTCTACACACCGCTCTTTATCCT
TAATTGCTTTTTCTATTTAGCGGAATTGATCGTTCTCAACTTGGTCGCCATTGC
15 AGCTCCACAGGCGAAAAAATCGGTGGAAATGCCAATACAGGTGCACGGCGAG
TGCCGATAAGCTGGAAAAATCGGGAAAACGCACGCCTACACATTCATTGCCAG
CATCGGCTTTGCCTTTTCGCTGTCGAGATTAGCATATTTCCACTTTTGGTTCGC
GCACAACACTANCTAAATTATTGNTTATTTTTTCCCCAACTGTGAGGTGAAAC
TGTGAAACAAAACCACTGTGGGCGGGTCAGTGTGACCCTCTCGCGGTGGGTG
20 AAAATCCTAGTGAGCTTCGTTGTTAGGGCTGTATGACACGAAAGCAAGTTGAA
AAGAACTTTTTTAAAATTATTTGGTTAATTGAGCAGAACTAAACTATATN
AAAATATTTAAGAATNCAGATTAGTGATGTATTTAATAATAATAGTAAGAT
GTTC

25 **Rescue ID** EcoR1

Rescue Sequence 2

CTTNTTTGATAGANATAGGCTTCTTTTAAAAAAAANAAGCAGCANCAGGGG
CCCNAAAGTGCGTGNNTGTGAACGCTGATTGCTTGCAAGTGTGTTCTGTGTG
TGTGATTGTGTGCTCCGANCAAGTGAAATCAATAATTTGCAGCCACAAGCA
30 ATTAATAAAAACTGCAATAATGTCAAAAAATCTAATTGAGGCAACAAATTAN
CAAAGCCATNAAAGCAGGCTGCACTGCGAGAAAATTGTGCCTTTCCACAGAT
CTTCTGCTGCAAAGCNAAAGAANGTAAGCAAGTCGGCCANTTTATTNCATTCT
TCTCATCTCTCTTCTTCGCGAATTGGCGCNTANCACTTACAATAATTNATATNA
CTTCTTAAATTTCAAANTCCCTTTTNTGAACGGANCTTTTAACGGAAAACAAA
35 GCGGGTAAACTAACTTAACTAACTAATTANAANTGTANGTATAAATGAACC
GAACTCGCTTTAGATATNATGCGTTTCACTAACANATTANAACAACTTTGAA
GCTGTANTGTCAGGTTGTTATTNCGTTCACCANATGTAGACTGNCCGNNAATT
TNACCTTTCCCATANTCTGTTCTTAANTGTNTTGTTTTTTCCCAATNNTTGTATC
ATNCNTTGGTNAATNANCTNAACGGCCCAAAGTNAATGAATTCCANTCACGTC
40 CACTGGCTCTGGTTCNATANTTAATNGGCTGTTTCTTACTTCCCTTAACCCTAA
CATCTNNTAATCACCTGTGCCATNTGTTTGTGTGTGTGAACGAATGAGAAA
AAAAA

Annotated *Drosophila* genome genomic segment

AE003825

Annotated *Drosophila* genome Complete gene candidate CG9005 - novel putative cell adhesion

5 **Human homologue of Complete gene candidate** CG9005- Ensembl predicted gene
ENSP00000006008
Gene:ENSG00000005238
Clone:AC004472
Contig:AC004472.00001 6.00E-38
10 (KIAA1539 protein AB040972) and
AK022837 Homo sapiens cDNA
FLJ12775 4e-33

15 **Putative function** Putative cell adhesion protein

Confirmation by RNAi Reduced G2/M peak

Example 67 (Category 5)

	Line ID	74/3
	Category	2nd chromosome, small imaginal discs
5	Reversion	NR
	Map Position	47A
	Rescue ID	EcoR1
	Rescue Sequence	
10	GCACAGAATGGCNCCTTCACGACAAAAGATCTNCNAATTAGGATGATGCAGA AGGAGGACACGCTTTTCATTATCTGGTTGCCACCTAATTTAAGTTCCACATCAA GGAAGAAGGAAATACGTTCCAACGGACGTCAAATTTACTAACTACACTACTT GAAAAGCCTGTCTATAAAAAACACGATAACGTTTTTGTCTAATCTCAAGACAATG TTAAATATAATTGGAGAAAGTATTGAATATGAATATCACAAAAATTGTTTAGG 15 GTCTCTACGTGGTAAATAGTATTTGGCATAGACAGTGAGATGTGAGTCGTACG TACTAATTAATAAAGTTGTTCAARAGAACCTCATATACTGTAAGTGACAACGA ACGAAGCTGACAACTCTGCTTGACATATTTGGCGGAGTTCGAAAATATCATC GCATTGGTATTGTTTTTGTNTCCACCNTGGGGCGAGATTTTGTGTTGCTTTAC TTTGCTTGTTTTTTCNCCACAAANCGAACCATATGTTTCGAAATGGTAAATTA 20 CCGTGCCAACAAGCTCTCTCTCTCCCCACTCCGAAACTCTCTCATCTCTCCTTG CAATTGTTTAAGGTGTGCAAGGAAATGAAAAATGTCCCGGCTGTGTTNCCATG CATTCCCCTTCAAAGCCAATTATNTTGTGCCTCTCCAACNTTTTGATCGGNN TGATTTTTTTGGCTCCCCNTANTCCCCCCCCCTTCNCCATTCCGGGTTANAT TATTNTNCCAATTTTCCTATTTTACGGTCCCGTCCCTGGAAATANTTCCTNC 25 AATCNCCGCTCCATNTCNCCATNTTTGACAGATTTTC	
	Annotated <i>Drosophila</i> genome genomic segment	AE003829
30	Annotated <i>Drosophila</i> genome Complete gene candidate	CG12052 lola -a specific RNA polymerase II transcription factor involved in axon guidance
35	Human homologue of Complete gene candidate	1e-09 3789797 (AF059569) actin binding protein MAYVEN [Homo sapiens]
	Putative function	lola-like specific RNA polymerase II transcription factor
40	Confirmation by RNAi	Almost no G1 peak and increase in G2/M peak indicating arrest in G2/M

Example 68 (Category 5)

Line ID 79/7
Category 2nd chromosome, small imaginal discs
5 **Reversion** R
Map Position 55B

Rescue ID BamH1

Rescue Sequence 1

10 GTCTCATGCACCCTGGCCCTNAGCTGCATAAGTGTAAGTGTGTGNCTGTGTGC
GAGTGTGGGTAGGCGGCGGCAACTATCTCGCTTGCTCTTGCCTCCGGGGTTAT
CGGTAGCTTCTTCTAGGCTGAGTGCATTTCGTTGAATCGTGGATGTTGAAAGTT
GTCTAATTTCCGAACATTGATTTTTCCCTTCCCGTCAAGAACTGCATTGT
TGCTTCTTGAAGACCAGTTTTGGTAACATCAGGAGAATGGAAAGGAGCGAGT
15 GAGTCGGTGAGTAAGTGAGTGAGCGATGCGAGCGACAAAATCAACAACAACA
ACAACAACGGTCAAAACGAGTTCCAACGAAAGTTGCAACACTCTCAACAATT
TGAGCAGCTCCGTTTGTGTTATTGCATTACTCAATCGGGAAGACTCTACACTC
GACGGAATAGTGTGCTCGTCTGAAATTTATCNATTTCCATTCCCTTCCTTTGTTT
TTGGGCCAAACAATGGCNTCGGCAANCGTTCGTGGAAAACCGCAGGAACCAC
20 CAAAATGCCTGGCGTCACATTAACCGAGCCGCCTTTGTTTATGCAAAATATTATT
GTAATATTTGGTNAAAATTAAGTCGCGCTTCNCGTTACTTTTTATTTCATATAC
ACGCAGCAGCAGCACGCATACAGTCACGTCACGCACACATACAATCGCCGTN
CACATACACTTGTCTTTTTNCCACACACTTTCCTAATCAT

25 **Rescue ID** EcoR1

Rescue Sequence 2

NGGNGTCTCATGCACCCTGGCCCTNAGCTGCATAAGTGTAAGTGTGTGNCTGT
GTGCGAGTGTGGGTAGGCGGCGGCAACTATCTCGCTTGCTCTTGCCTCCGGGG
TTATCGGTAGCTTCTTCTAGGCTGAGTGCATTTCGTTGAATCGTGGATGTTGAA
30 AGTTGTCTAATTTCCGAACATTGATTTTTCCCTTCCCGTCAAGAACTGCA
TTGTTGCTTCTTGAAGACCAGTTTTGGTAACATCAGGAGAATGGAAAGGAGCG
AGTGAGTCGGTGAGTAAGTGAGTGAGCGATGCGAGCGACAAAATCAACAACA
ACAACAACAACGGTTCAAAACGAGTTCCAACGAAAGTTGCAACACTCTCAAC
AATTTGAGCAGCTCCGTTTGTGTTATTGCATTACTCAATCGGGAAGAACTCTA
35 CACTCGACGGAATAGTGTGCTCGTCTGAAATTTATCNATTTCCATTCCCTTCCTT
TGTTTTTGGGCCAAACAATGGCNTCGGCAANCGTTCGTGGAAAACCGCAGGA
ACCACCAAAATGCCTGGCGTCACATTAACCGAGCCGCCTTTGTTTATGCAAAAT
ATTATTGTAATATTTGGTNAAAATTAAGTCGCGCTTCNCGTTACTTTTTATTTC
ATATACACGCAGCAGCACGCATACAGTCACGTCACGCACACATACAATCGCC
40 GTNCACATACACTTGTCTTTTTNCCACACACTTTCCTAATCATNNTA

Genomic hit, Accession No. AC004296

Associated ORF

45 Genscan: ORF2 predicted sequences >15:31:31|GENSCAN_predicted_peptide_3|109_aa
MVTSEFRHLRDEKSFTDVTLACEGQTCKAHKMWLSACSPYFKALLEENPSKHPHIL

KDVSYIHLQAILEFM YAGEVNVSQEQLPAFLKTADRLKVKGLAETPSSIKREG

>15:31:31|GENSCAN_predicted_CDS_3|330_bp

5 atggtgacctcggtccgtcacctgcgcgacgagaagagcttcacagatgtaacactgcctgcgagggccaaacctgcaaagcc
cacaaatgggtgcttccgcttcagtcctactttaagcgctactggaggagaacccatcgaagcatccgatcattatcctgaaa
gatgtctcctacattcacctacaggctatactggagttcatgtacgccggtgaggtgaacgtgtcccaggaacaattgccagcatt
cttaagaccgccgatcgctcaaagtgaaggcctcgagagacaccagttcgataaagcggaaggtga

10 **Drosophila Gene Hit** TBLASTN with ORF2: several zinc finger proteins including
Broad-Complex mRNA for BRcore-Z2 protein (X54665)
Human Homologue TBLASTN with ORF2: kelch (*Drosophila*)-like 2 (Mayven actin
binding protein) (KLHL2) (AF059569)

15 **Annotated Drosophila genome genomic segment** AE003800
Annotated Drosophila genome Complete gene candidate CG5738- lola, lola-like
putative kelch-like putative
specific RNA polymerase II
transcription factor known to
affect disc morphology

20 or could be CG10914 - novel
unknown

25 **Human homologue of Complete gene candidate** CG5738- 9e-09 3789797
(AF059569) actin binding
protein MAYVEN [Homo
sapiens]

30 CG10914- predicted gene
ENSP00000051207
Gene:ENSG00000047313
Clone:AC068261
Contig:AC068261.00019
4.00E-49 (potential cell
35 division GTP binding protein
1: ENST00000051207

40 **Putative function** CG5738: lola like specific RNA polymerase II transcription factor,
CG10914: Possible GTP binding protein

Confirmation by RNAi Both show marked reduction in G1 to G2/M ratio

Example 69 (Category 5)

Line ID 80/2, 81/8
Category 2nd chromosome, small imaginal discs
5 **Reversion** R
Map Position 57D/E

Rescue ID BamH1

Rescue Sequence 1

10 CANTTTCAGAGGCCATAGNCCTTCACAAAATTCNCCATCTCTGCCCCGGCATCC
GTGCTTGAAAATGGTGCCAATGCGTCGTGGAGAATCTGCTGCACTCGATGGTC
TGCAAAATTGCACATTTATTAGATTTAATAAAATTTTCAACTGTCCGCGANCAC
GTTTGCTCGTGTTGAATTTTCGAGTACAAAATTAGTGCGACTGTTGGATTGCATT
GAAATGCCAAAAATCGGTGTGACCATTTTGAAGTCCCCACAGGCTCATGACTT
15 TCGCGGTTACCAAATCCAAATAACGCAAGCTGGTCACGCTGTCAAACATCGG
TGACGGAATGGTGACGACACAAACAATTTGCTTAAAAACTTTCTTGCGGCCGT
AAAAATGCGCAAGCAGCCTGGCAGCGCAACGCACGTACACGTAATTGGAACA
AATGTTTGCTGAACCACAACCGCCCACTAAATGTTANCCGCCAAGTCTTTTCC
CCCGCCGCCCGCGTCNTCNCNCCGGATTATTTGGTTTACAATTTGCTTAC
20 ACAAGTGCAATCGTCGATAGCGCTTCATTTGGAGTAACAAGTAATATTTTGC
GCCGTACTGCTGTTTCGCCGTATCAGACAGAAGGTTGGTATCAGTTCGACGCAG
CTTGTGACGGTATTGCATACGCGGCGAAACGCCACGTGAAAACGGATCGCA
GTTCTCGAAAACCTCNGGATAAAAA

25 **Rescue ID** EcoR1

Rescue Sequence 2

TGGGGTCTCANGCCCCGACGGCCATATTTTAACACAAGATTCNNCANCTCTGC
AGGGCATCCGTGCTTGAAAATGGTGCCAATGCGTCGTGGAGAATCTGCTGCAC
TCGATGGTCTGCAAAATTGCACATTTATTAGATTTAATAAAATTTTCAACTGTC
30 CGCGAGCACGTTTGCTCGGTGTTGAATTTTCGAGTACAAAATTAGTGCGACTGT
TGGATTGCATTGAAATGCCAAAAATCGGTGTGACCATTTTGAAGTCCCCACAG
GCTCATGACTTTTCGCGGTTACCAAATCCAAATAACGCAAGCTGGTCACGCTG
TCAAACATCGGTGACGGAATGGTGACGACACAAACAATTTGCTTAAAAACTTT
CTTGCGGCCGTAAAAATGCGCAAGCAGCCTGGCAGCGCAACGCACGTACACG
35 TAATTGGAACAAATGTTTGCTGAACCACAACCGCCCACTAAATGTTAGCGCCA
ACTNCTTTTCCCCGCCCGCCGGTCGTCNTCNCNCCGGATTATTTTGGTTTACA
ATTTGCTTACACAAGTGCAATCGTCGATAGCGCTTCATTTTGGAGTAACAAGT
AGTATTTTGCGCCGTACTGCTGTTTCGCCGTATCANACAGAAGGTTGGTATCAG
TTCGACGCAGCTTGTGACGGTATGCATACGCGGGGAAACGCCACGTGAAAAC
40 GGATCGCAGTNCTCGAAACTCNGGATAAAAGAAAAAGTAGGCTGAATG

Genomic hit, Accession No. AC007175

Associated ORF

45 Genscan: ORF2 predicted sequences >16:09:09|GENSCAN_predicted_peptide_3|2497_aa
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Drosophila Gene Hit TBLASTN with ORF2: brahma protein (M85049) and imitation-SWI protein (ISWI) (L27127) and chromodomain-helicase-DNA-binding (CHD-1)

45 **Human Homologue** BLASTX with EST TBLASTN with ORF2: Snf2-related CBP activator protein (SRCAP) (AF143946) and SWI/SNF related, matrix associated, actin dependent regulator of chromatin, subfamily a, member 4 (SMARCA4) (NM_003072.1)

	<i>Drosophila</i> EST	several including SD07794 (AI534784), LD34465 (AA990657)
	Annotated <i>Drosophila</i> genome genomic segment	AE003453
5	Annotated <i>Drosophila</i> genome Complete gene candidate	CG9696 – domino an enzyme involved in DNA repair homology to snf2 family helicases
10	Human homologue of Complete gene candidate	CG9696- gi4557447 416409C913D6A935 [refNP_001261.1] chromodomain helicase DNA binding protein 1 [Homo sapiens] (1.90E-85
15	Putative function	snf2 helicase family member protein that contains a chromodomain, which occurs in proteins that are implicated in chromatin compaction, and an SNF2/SWI2-like helicase domain, which occurs in proteins 20 that are believed to activate transcription by counteracting the repressive effects of chromatin structure
25	Confirmation by RNAi	Loss of G1, peak, increase in G2M indicating arrest in G2/M

Example 70 (Category 5)

Line ID 99/31
Category 2nd chromosome, small imaginal discs
5 **Reversion** NR
Map Position 53E

Rescue ID EcoR1

Rescue Sequence 1

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Rescue ID BamH1

Rescue Sequence 2

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Genomic hit, Accession No. CSC:AC020063

45

Associated ORF

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 40 ctaacaacaagtcaaaaccagctggaaaggccgatagtcgcgaaaaaatcaaaagaaggaatcgtccgagggaagatgatgat
 gtcgatgacaaagatgaatccgacgaggatgagccactaaccaaaaggcgaacaggcattcccaacggatgaacaaatac
 cggatattgcaagagattctgataaagccaatcttgaggagattacgatgaaaccgtgtgcaacaagtttatgcaaaatattcc
 agactttgacctaacagacaagaagactcatcaagcgcagctgaaagcggacggagttcaggatttgatgtagtcccga
 actgatccgcgtggccgaacaacggttacaatatggtgatctgtgtgcaacaatcagatattggggagacgtaa

45 Human Homologue TBLASTN with ORF1: poor homology with DEK gene
 (D6S231E) (NM_003472.1)
Drosophila EST several including LD33301 (AA979048)

	Annotated <i>Drosophila</i> genome genomic segment	AE003805
	Annotated <i>Drosophila</i> genome Complete gene candidate	CG5935 - EG:EG0003.6 - novel with weak homology to DEK oncogene
5		CG8648 - EG:EG0003.3 - novel XPG/ flap endonuclease-like, DNA repair?
10	Human homologue of Complete gene candidate	CG5935- 1e-17 4503249 ref NP_003463.1 pD6S231E DEK gene >gi 544150 sp P35659 DEK_H UMAN DEK PROTEIN >gi 284375
15		CG8648- 4758356 ref NP_004102.1 pFEN1 flap structure-specific endonuclease 1;
20		MATURATION FACTOR 1 (MF1); DNase IV; RAD2_HUMAN(aa)
25	Putative function	CG5935: function unknown but putative DNA-binding protein predicted to be involved in chromosomal organisation. The translocation (6;9), associated with a specific subtype of acute myeloid leukemia, results in the fusion of two genes, dek and can, and the expression of a chimeric, leukemia-specific dek-can mRNA
30		CG8648: Novel XPG/ flap endonuclease-like, DNA repair protein
	Confirmation by RNAi	Both show slight reduction of G1 peak

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Each of the applications and patents mentioned above, and each document cited or referenced in each of the foregoing applications and patents, including during the prosecution of each of the foregoing applications and patents ("application cited documents") and any manufacturer's instructions or catalogues for any products cited or mentioned in each of the foregoing applications and patents and in any of the application cited documents, are hereby incorporated herein by reference. Furthermore, all documents cited in this text, and all documents cited or referenced in documents cited in this text, and any manufacturer's instructions or catalogues for any products cited or mentioned in this text, are hereby incorporated herein by reference.

Various modifications and variations of the described methods and system of the invention will be apparent to those skilled in the art without departing from the scope and spirit of the invention. Although the invention has been described in connection with specific preferred embodiments, it should be understood that the invention as claimed should not be unduly limited to such specific embodiments. Indeed, various modifications of the described modes for carrying out the invention which are obvious to those skilled in molecular biology or related fields are intended to be within the scope of the following claims.

CLAIMS

1. A polynucleotide selected from:
 - (a) polynucleotides comprising any one of the nucleotide sequences set out in Examples 1 to 70 or the complement thereof.
 - 5 (b) polynucleotides comprising a nucleotide sequence capable of hybridising to the nucleotide sequences set out in Examples 1 to 70, or a fragment thereof.
 - (c) polynucleotides comprising a nucleotide sequence capable of hybridising to the complement of the nucleotide sequences set out in Examples 1 to 70 or a fragment thereof.
 - 10 (d) polynucleotides comprising a polynucleotide sequence which is degenerate as a result of the genetic code to the polynucleotides defined in (a), (b) or (c).
2. A polynucleotide selected from:
 - (a) polynucleotides comprising any one of the nucleotide sequences set out in Examples 1 to 14 or the complement thereof.
 - 15 (b) polynucleotides comprising a nucleotide sequence capable of hybridising to the nucleotide sequences set out in Examples 1 to 14, or a fragment thereof.
 - (c) polynucleotides comprising a nucleotide sequence capable of hybridising to the complement of the nucleotide sequences set out in Examples 1 to 14 or a fragment thereof.
 - 20 (d) polynucleotides comprising a polynucleotide sequence which is degenerate as a result of the genetic code to the polynucleotides defined in (a), (b) or (c).
3. A polynucleotide selected from:

- (a) polynucleotides comprising any one of the nucleotide sequences set out in Examples 15 to 19 or the complement thereof.
- (b) polynucleotides comprising a nucleotide sequence capable of hybridising to the nucleotide sequences set out in Examples 15 to 19, or a fragment thereof.
- 5 (c) polynucleotides comprising a nucleotide sequence capable of hybridising to the complement of the nucleotide sequences set out in Examples 15 to 19 or a fragment thereof.
- (d) polynucleotides comprising a polynucleotide sequence which is degenerate as a result of the genetic code to the polynucleotides defined in (a), (b) or (c).
- 10 4. A polynucleotide selected from:
 - (a) polynucleotides comprising any one of the nucleotide sequences set out in Examples 20 to 30 or the complement thereof.
 - (b) polynucleotides comprising a nucleotide sequence capable of hybridising to the nucleotide sequences set out in Examples 20 to 30, or a fragment thereof.
 - 15 (c) polynucleotides comprising a nucleotide sequence capable of hybridising to the complement of the nucleotide sequences set out in Examples 20 to 30 or a fragment thereof.
 - (d) polynucleotides comprising a polynucleotide sequence which is degenerate as a result of the genetic code to the polynucleotides defined in (a), (b) or (c).
- 20 5. A polynucleotide selected from:
 - (a) polynucleotides comprising any one of the nucleotide sequences set out in Examples 31 to 53 or the complement thereof.
 - (b) polynucleotides comprising a nucleotide sequence capable of hybridising to the nucleotide sequences set out in Examples 31 to 53, or a fragment thereof.

- (c) polynucleotides comprising a nucleotide sequence capable of hybridising to the complement of the nucleotide sequences set out in 31 to 53 or a fragment thereof.
- (d) polynucleotides comprising a polynucleotide sequence which is degenerate as a result of the genetic code to the polynucleotides defined in (a), (b) or (c).
- 5
6. A polynucleotide selected from:
- (a) polynucleotides comprising any one of the nucleotide sequences set out in 54 to 70 or the complement thereof.
- (b) polynucleotides comprising a nucleotide sequence capable of hybridising to the nucleotide sequences set out in 54 to 70, or a fragment thereof.
- 10
- (c) polynucleotides comprising a nucleotide sequence capable of hybridising to the complement of the nucleotide sequences set out in 54 to 70 or a fragment thereof.
- (d) polynucleotides comprising a polynucleotide sequence which is degenerate as a result of the genetic code to the polynucleotides defined in (a), (b) or (c).
- 15
7. A polynucleotide probe which comprises a fragment of at least 15 nucleotides of a polynucleotide according to any of Claims 1 to 6.
8. A polypeptide which comprises any one of the amino acid sequences set out in Examples 1 to 70 or in any of Examples 1 to 14, Examples 15 to 19, Examples 20 to 30, Examples 31 to 53 and Examples 54 to 70, or a homologue, variant, derivative or fragment thereof.
- 20
9. A polynucleotide encoding a polypeptide according to Claim 8.
10. A vector comprising a polynucleotide according to any of Claims 1 to 7 and 9.

11. An expression vector comprising a polynucleotide according to any of Claims 1 to 7 and 9 operably linked to a regulatory sequence capable of directing expression of said polynucleotide in a host cell.
12. An antibody capable of binding a polypeptide according to Claim 8.
- 5 13. A method for detecting the presence or absence of a polynucleotide according to any of Claims 1 to 7 and 9 in a biological sample which comprises:
- (a) bringing the biological sample containing DNA or RNA into contact with a probe according to Claim 9 under hybridising conditions; and
 - (b) detecting any duplex formed between the probe and nucleic acid in the
- 10 sample.
14. A method for detecting a polypeptide according to Claim 8 present in a biological sample which comprises:
- (a) providing an antibody according to Claim 12;
 - (b) incubating a biological sample with said antibody under conditions which
- 15 allow for the formation of an antibody-antigen complex; and
- (c) determining whether antibody-antigen complex comprising said antibody is formed.
15. A polynucleotide according to according to any of Claims 1 to 7 and 9 for use in therapy.
- 20 16. A polypeptide according to Claim 8 for use in therapy.
17. An antibody according to Claim 12 for use in therapy.

18. A method of treating a tumour or a patient suffering from a proliferative disease comprising administering to a patient in need of treatment an effective amount of a polynucleotide according to any of Claims 1 to 7 and 9.
19. A method of treating a tumour or a patient suffering from a proliferative disease,
5 comprising administering to a patient in need of treatment an effective amount of a polypeptide according to Claim 8.
20. A method of treating a tumour or a patient suffering from a proliferative disease, comprising administering to a patient in need of treatment an effective amount of an antibody according to Claim 12 to a patient.
- 10 21. Use of a polypeptide according to Claim 8 in a method of identifying a substance capable of affecting the function of the corresponding gene.
22. Use of a polypeptide according to Claim 8 in an assay for identifying a substance capable of inhibiting the cell division cycle.
23. Use as claimed in Claim 22, in which the substance is capable of inhibiting mitosis
15 and/or meiosis.
24. A method for identifying a substance capable of binding to a polypeptide according to Claim 8, which method comprises incubating the polypeptide with a candidate substance under suitable conditions and determining whether the substance binds to the polypeptide.
- 20 25. A method for identifying a substance capable of modulating the function of a polypeptide according to Claim 8 or a polypeptide encoded by a polynucleotide according to any of Claims 1 to 7 and 9, the method comprising the steps of: incubating the polypeptide with a candidate substance and determining whether activity of the polypeptide is thereby modulated.

26. A substance identified by a method or assay according to any of Claims 21 to 25.
27. Use of a substance according to Claim 26 in a method of inhibiting the function of a polypeptide.
28. Use of a substance according to Claim 26 in a method of regulating a cell division
5 cycle function.